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THE VASCULAR ANATOMY OF DIMEROUS AND TRIMEROUS SEEDLINGS OF PHASEOLUS VULGARIS

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INTRODUCTORY

The great majority of investigations dealing with the anatomy of plants have been purely descriptive in character. As a result of observation, the typical or average condition of plant structures has been recorded in terms which are general and often indefinite. Comparatively few morphological papers deal with the problem of the variation of the structures under consideration, treat of their correlations with one another, or even present the detailed measurements which might serve for the solution of such fundamental morphological problems.

The older comparative morphology is indispensable. It provides a general knowledge of plant structures and serves as a basis for the classification of the vegetable kingdom. The recognition that description must be supplemented by the results of experimentation has, however, led to the establishment of the newer special science of experimental morphology. The time has come to extend still further our study of plant form by calling to the service of vegetable morphology the methods of measurement and mathematical analysis. These methods are particularly useful in an attack upon the fundamental problems of morphogenesis. It is by measuring exactly the various plant structures during their successive stages of development, in terms of size or number; by determining their relative variability in different organs or regions of the plant, or under varying external conditions, and by discovering such correlations as exist, both among the structures themselves and between them and their progenitors and their environment, that we shall be able to build up a body of fact on which morphogenetic theory may rest.

The present paper gives a portion of the results of a biometric analysis of a comparatively simple morphological problem, that of the gross vascular anatomy of certain normal and abnormal bean seedlings. Our purpose has been:

1. A study of the vascular anatomy of normal and of abnormal seedlings from the point of view of descriptive morphology—a preliminary which we believe to be essential to a sound interpretation of any statistical results.
2. A statistical study of the number and variation of the vascular elements in different regions of the seedling.

3. An investigation of the correlations between these internal characters (such as those which exist between bundle number in different regions of the seedling) and between the internal characters and external features of the plant.

The results of the first and second phases of the investigation are set forth in the present paper; the third is reserved for a later publication.

MATERIALS AND METHODS

A priori considerations seemed to indicate that a promising line of attack upon the general field of quantitative plant morphology lay in the investigation of vascular bundle number. Such an investigation should be on a scale sufficiently large to make possible the determination of trustworthy biometric constants, and should have as its subject a plant organ of relatively simple but variable structure. Because of the ease with which they can be grown in quantity, their sharply marked external characteristics, their convenient size for histological work, and their relatively simple internal structure, seedlings of *Phaseolus vulgaris* furnish highly satisfactory material for a study of variation and correlation in vascular structures.

Among the many types of variant seedlings of the garden beans which may be secured by extensive plantings, two were selected for investigation: (a) normal (*dimerous*) seedlings, with two cotyledons and two primordial leaves, and (b) *trimerous* seedlings, with three cotyledons and three primordial leaves. For brevity in table headings the dimerous plants will sometimes be represented by "2-2" and the trimerous by "3-3," where the first figure gives the number of cotyledons and the second the number of primordial leaves.

Since one of the purposes of this work is to carry out a comparison of bundle number in normal and teratological seedlings, the selection of a satisfactory control series of normal plants is a matter of primary importance. It is essential that the seedlings of the types to be compared be selected in a manner to reduce to a minimum any external influences tending to bring about differences between them. It is clear that if the abnormal and the normal seedlings were taken from different series of parent plants, either genetic differences or environmental influences acting upon the parent plant might be effective in bringing about a differentiation in the characters of the seedling examined. A normal seedling from the same parent was, therefore, taken for comparison with each abnormal seedling¹ in each series in which the seed was derived from individual parent plants. Closer control of the influence of innate differences in the parents and of the possible influence of parental environment hardly seems practicable since the

¹ In the vast majority of the cases one abnormal seedling only was sectioned from a parent plant. When more than one abnormal seedling was available a control was taken for each. Naturally it is immaterial whether control *a* or *b* be compared with abnormal seedling *A* or *B*, since all are siblings.

pairs of abnormal and normal seedlings were, in three of the lines investigated, derived from the same parent plant.

Furthermore, care was taken that seedlings compared were grown under essentially identical conditions, in order to reduce to a minimum the environmental influences which might possibly tend to bring about differences between them. Seeds from individual plants were germinated in flats and harvested as soon as possible after they broke through the sand. Thus all seeds not only developed under the same parental environment but were germinated under sensibly identical conditions, were collected simultaneously, and were in consequence sectioned at essentially the same stage of maturity.

Because of the rapidity with which seedlings change and the great influence of temperature upon growth, it is difficult to standardize, or exactly to describe, the stage of development at which the seedlings were taken. Most of them were placed in alcohol before or very soon after the primordial leaves had unfolded. Thus a fairly uniform and early stage of development was secured.²

Free-hand sections were cut and mounted temporarily. When necessary, phloroglucin and hydrochloric acid were used to bring out the vascular bundles. The general vascular topography of the seedlings was studied, but the data for the statistical analysis of the seedling anatomy were derived from a careful count of the number of vascular bundles at various levels in the seedling. Because of a certain amount of variation in the number of bundles with position in the organ, counts were made in definite regions only—the basal region of the hypocotyl (just at the point of transition from “root structure” to “stem structure”); the median region of the hypocotyl; and the median region of the epicotyl. In three series counts were also made of the protoxylem poles in the upper portion of the primary root.

The number of data available for the several regions differs because of a change in the plan of the work. Sectioning and counting were begun by two of us at Cold Spring Harbor in the summer of 1917 and continued with the assistance of Miss Eunice Kinnear in the summer of 1918. This work was confined to the mid-regions of the hypocotyl and epicotyl. From a statistical study of these data it seemed desirable to have a further series of countings made independently by a specialist in vascular anatomy. The work was, therefore, continued at Storrs during 1918, 1919, and 1920. We are greatly indebted to Miss Flora Miller for assistance in this phase of the work. At Storrs, sections were made at the base of the hypocotyl as well as in the mid-region of hypocotyl and epicotyl. In three series, sections were made of the root as well.

The bundles vary considerably in size, the largest being well developed

² Some of the seedlings of line 143 were allowed to become a little older, but there is no evidence of change in bundle number with age.

and the smallest containing only one or two lignified xylem cells and a small patch of phloem. Some are even more reduced, consisting of a phloem patch alone. Any strand in which at least one well lignified xylem element could be made out was counted as a bundle. Some of the bundles are partially double in character, this condition being due either to partial fusion or to incipient division. Whenever such a strand was surrounded by one bundle sheath it was counted as one bundle; when the separation was so great that the bundle sheath itself showed signs of division, the strand was counted as two.

The seedlings were harvested at a stage when the vascular tissues of the first epicotyledonary internode were not completely lignified, and the number of bundles counted was therefore possibly less than the number which would finally be developed there.

None of these possible sources of error is believed to be great enough to affect the conclusions appreciably.

THE STRUCTURE OF THE SEEDLING

In order to provide a sound basis for the understanding and interpretation of our later work, it is necessary to present a brief descriptive account of the structure of the seedlings.

The Normal (Dimerous) Seedling

The morphology of the seedling of *Phaseolus* has received the attention of several investigators, notably Dodel³ and Compton.⁴ Like most of the large seedlings of the Leguminosae it is normally tetrarch in fundamental plan; that is, there are four groups of protoxylem elements in the root. At a very early stage there is associated with each of these a group of metaxylem cells. It is these groups of metaxylem elements, throughout the whole seedling, which in the present paper are counted as "bundles," even though (as is sometimes the case) they are not associated with protoxylem clusters.

At the stage when these seedlings were harvested, cambial activity had hardly begun to show itself, so that these primary bundles remained distinct and easy to identify.

The condition in the upper part of the root of a normal seedling is shown in figure 1. The four bundles, two in the cotyledonary plane and two in the intercotyledonary plane, are more or less V-shaped (with the protoxylem group in an exarch position at the apex of the V) and tend to extend laterally. They surround a large pith. In passing up into the base of the hypocotyl, each of these bundles divides into two (fig. 2), and typical stem structure,

³ Dodel, A. Der Übergang des Dicotyledonen-stengels in die Pfahl-wurzel. Pringsh. Jahrb. 8: 149-193. 1872.

⁴ Compton, R. H. An investigation of the seedling structure in the Leguminosae. Jour. Linn. Soc. 41: 7-122. 1912.

with the protoxylem in an endarch position, begins to be assumed. Each pair is subsequently referred to as a "primary double bundle." Thus the level of transition from root structure to stem structure is low, being prac-

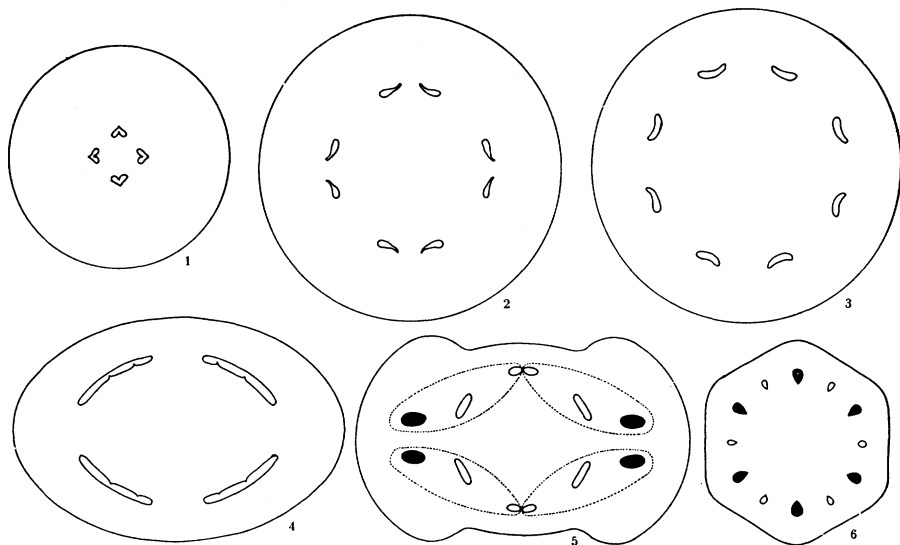


FIG. 1. Dimerous seedling. Transverse section through the root, showing its tetrarch condition (four protoxylem poles). FIG. 2. Dimerous seedling. Transverse section through the base of the hypocotyl showing the four primary double bundles, each of which has been derived from one of the four root strands. FIG. 3. Dimerous seedling. Transverse section through the mid-region of the hypocotyl showing the normal eight-bundled condition. No intercalary bundles are figured. FIG. 4. Dimerous seedling. Transverse section just below the cotyledonary node. The four bundles or bundle groups have originated by a more or less complete fusion of the adjacent members of each of the original pairs. Each bundle, as shown by the two constrictions in it, is about to break up into the three strands shown in figure 5. FIG. 5. Dimerous seedling. Transverse section through the cotyledonary node. Each group of three strands which have arisen by a breaking up of the large bundles in figure 4 is here enclosed by a dotted line. These three strands are a cotyledonary trace (solid black), an epicotyledonary bundle, and a small bundle which will fuse with its adjacent neighbor to form another epicotyledonary bundle. FIG. 6. Dimerous seedling. Transverse section through the mid-region of the epicotyl showing the twelve bundles which have arisen by the splitting of the six original epicotyledonary bundles. The six strands which are to go off as traces to the two primordial leaves are solid black.

tically at the base of the hypocotyl. The members of each of these four pairs soon separate until the eight bundles are approximately equidistant (fig. 3), a condition which persists throughout the hypocotyl until the cotyledonary node is approached.

In addition to these bundles, there are in a considerable percentage of the normal seedlings studied a variable number of accessory or intercalary bundles, the "Zwischenstränge" of Dodel. These may make their appear-

ance in the upper part of the root or in the lower region of the hypocotyl, some ending blindly below and others arising by division of the primary bundles. These intercalary bundles, which are not a very common feature of seedling anatomy in general, perhaps serve to increase the conductive capacity of the hypocotyl and may be associated with the large size of the seedling. They usually lack protoxylem elements.

At the cotyledonary node there is a rather complex anastomosis of the bundle system. The details of this vary somewhat, but its fundamental features are as follows: The two members of each of the two original pairs of bundles in the cotyledonary plane (that is, opposite the two points where the cotyledons will later arise) become widely separated, and each member fuses with the adjacent member of the intercotyledonary pair (fig. 4). Four large bundles or bundle aggregates are thus produced. Each breaks up immediately, usually into three parts. The lateral member of each group of three which is in the *cotyledonary* plane approaches the corresponding bundle of the next group of three, and these two strands become the cotyledonary traces and enter the base of the cotyledon. The lateral member of each group of three which is in the *intercotyledonary* plane approaches the corresponding bundle of the next group and fuses with it. The changes which are made and the resultant condition at this stage are shown in figure 5. Two strands (solid black) are here departing to each cotyledon, and six bundles are left as the basis for the vascular system of the epicotyl. The details of this nodal complex vary somewhat owing to the different levels at which fusion and separation of bundles take place, and to the presence of intercalary bundles. These intercalary bundles, as they approach the cotyledonary node, fuse with the others and are completely lost, exactly six epicotyledonary strands almost invariably emerging from the complex, quite regardless of the number of intercalary bundles which may have entered it from the hypocotyl. This fact we shall find to be of importance when we consider the statistical relationships of bundle number in hypocotyl and epicotyl.

Above the cotyledons, the six remaining bundles approach one another, closing the cotyledonary gaps and forming a ring, the members of which almost immediately divide. The twelve bundles thus produced (fig. 6) persist throughout the first internode of the epicotyl.

At the first node of the epicotyl are inserted the two primordial leaves. *Phaseolus*, like other Leguminosae which have been investigated, possesses a trilacunar node, the leaf being supplied by three traces, each of which causes a separate gap in the vascular ring.⁵ The two primary leaves therefore remove six of the twelve bundles of the epicotyl (solid black in fig. 6). The six new bundles which appear just above the cotyledonary node are, therefore, evidently downwardly extending leaf traces. These facts make

⁵ Sinnott, E. W. The anatomy of the node as an aid in the classification of Angiosperms. *Amer. Jour. Bot.* 1: 303-322. 1914.

understandable the almost invariably twelve-bundled condition of the first epicotyledonary internode.

The structure of the normal seedling thus corresponds to the type found by one of the writers⁶ to be characteristic of a large number of Angiosperm families, in which the vascular supply to each cotyledon, consisting of two strands, leaves but one gap in the vascular ring; and in which the foliage leaf is trilacunar.

The Trimerous Seedling

The seedling with three cotyledons and three primordial leaves is built on a different plan from the normal one in that it is prevailingly hexarch, six

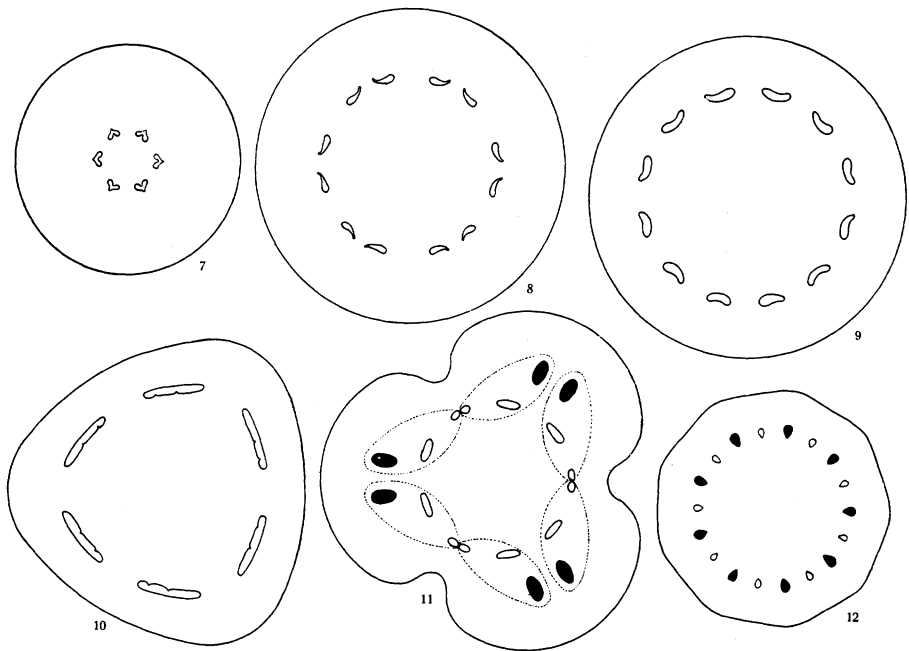


FIG. 7. Trimerous seedling. Transverse section through the root, showing its hexarch condition. FIG. 8. Trimerous seedling. Transverse section through the base of the hypocotyl, showing the six primary double bundles. FIG. 9. Trimerous seedling. Transverse section through the mid-region of the hypocotyl, showing the normal twelve-bundled condition. FIG. 10. Trimerous seedling. Transverse section just below the cotyledonary node. The six bundles or bundle groups correspond in origin and character to the four bundles of the dimerous seedling at this level. FIG. 11. Trimerous seedling. Transverse section through the cotyledonary node. Each group of three strands bounded by a dotted line corresponds in origin and character to a similar group at this level in the dimerous seedling. FIG. 12. Trimerous seedling. Transverse section through the mid-region of the epicotyl, showing the eighteen bundles which have arisen by the splitting of the nine original epicotyledonary bundles. The nine strands which are to go off as traces to the three primordial leaves are solid black.

⁶ Sinnott, E. W. Conservatism and variability in the seedling of dicotyledons. Amer. Jour. Bot. 5: 120-130. 1918.

bundles occurring in the upper part of the root (fig. 7). This number is soon reduced to five and eventually to four, in passing down the root.

Passing upward into the hypocotyl, the six main strands (the primary double bundles) divide to produce twelve (figs. 8 and 9). Intercalary bundles are much less common than in the normal seedlings, appearing in only a small percentage of cases, and then being rarely more than one or two in number. At the node the same general procedure is followed as in the normal seedling, except, of course, that there are more bundles concerned. Bundles of adjacent pairs approach and fuse (fig. 10). Each of these bundles or bundle aggregates then divides, generally into three. Three cotyledons are each supplied with two bundles (solid black), and three sets of three bundles each—each formed by the fusion of two lateral bundles in the intercotyledonary plane—remain behind. The bundle changes and the final condition at the departure of the cotyledonary traces are shown in figure 11. The epicotyledonary ring which forms from the bundles which remain thus consists of nine strands instead of the normal six. Many of these divide at once, although the number is not usually doubled, as in normal seedlings, but varies from 12 to 18 or even more in the mid-region of the epicotyl (fig. 12). The bundles are much more crowded than in the normal seedlings, which may perhaps account for the failure of some of them to divide at once.

A study of the first epicotyledonary node shows that three strands are given off to each primary leaf, leaving from 6 to 9 in the stem.

It is therefore evident that within classes of seedlings which are uniform externally there are considerable anatomical variations and that the two classes investigated are profoundly differentiated in their anatomical organization.

Our next task is to subject the mass of data upon which these general conclusions are based to a statistical analysis with the object of bringing out otherwise undeterminable relationships.

BUNDLE NUMBER AND ITS VARIATION AT DIFFERENT LEVELS IN THE SEEDLINGS

From the statistical side we have two problems to consider.

The first is that of the relative numbers of bundles at different levels, *i.e.*, in the root, at the base of the hypocotyl, in the central region of the hypocotyl, and in the epicotyl of the same plant in both normal and abnormal plants, together with the variability in bundle number in different regions.

The second is that of the differences in bundle number, and in variation of bundle number, between normal and abnormal plants.

Since it is impossible to consider type and variation of bundle number at different levels without noting differences in the trimerous and dimerous forms upon which the observations were based, we shall devote this section primarily to a parallel discussion of both problems.

We shall consider in order the levels at which sections were made, beginning at the root.

1. *Root.* Roots were sectioned in the cases of lines 93, 139, and 143. The numbers of bundles⁷ in the roots of normal and trimerous seedlings of these lines are shown in table 1.

TABLE 1

Primary Double Bundles	Line 93		Line 139		Line 143	
	Trimerous	Dimerous	Trimerous	Dimerous	Trimerous	Dimerous
3.....	—	—	2	—	4	—
4.....	31	132	15	149	37	219
5.....	87	20	53	1	113	2
6.....	34	—	36	—	66	—
7.....	—	—	—	—	1	—

The entries in this table show that most of the normal plants are tetrarch, although a small percentage are pentarch. In the trimerous seedlings the highest percentage are pentarch, but the remainder are distributed between tetrarch and hexarch with a few in more extreme classes. Sections made at progressively lower levels in the root show that the hexarch and pentarch conditions, in the trimerous seedlings, soon give way to tetrarch. This fact doubtless explains the relatively large number of non-hexarch cases

TABLE 2. *Vascular formula for base of hypocotyl of trimerous seedlings and their normal controls*

Base of Hypocotyl	Line 75		Line 93		Line 98		Line 139		Line 143	
	Trimerous	Dimerous	Trimerous	Dimerous	Trimerous	Dimerous	Trimerous	Dimerous	Trimerous	Dimerous
(4)	—	69	—	34	—	97	—	138	2	150
(4) + 1	—	30	—	37	—	43	1	9	3	55
(4) + 2	—	10	—	13	—	23	—	—	—	4
(4) + 3	—	4	—	5	—	2	—	—	—	—
(4) + 4	—	2	1	1	—	—	—	—	—	—
(4) + 5	—	2	—	—	1	—	—	—	—	—
(4) + 6	1	—	—	—	—	—	—	—	—	—
(5)	1	13	5	22	4	6	4	1	15	5
(5) + 1	8	4	10	18	6	8	4	2	31	5
(5) + 2	2	1	3	9	1	1	—	—	—	—
(5) + 3	—	1	—	1	—	1	—	—	—	—
(6)	107	5	120	10	160	1	92	—	134	—
(6) + 1	12	1	11	3	10	—	5	—	25	1
(6) + 2	2	—	1	2	—	—	—	—	—	—
(7)	7	—	4	—	1	—	—	—	5	1
(7) + 1	—	—	—	—	—	—	—	—	4	—
(7) + 2	1	—	—	—	—	—	—	—	—	—
(8)	1	—	—	—	—	—	—	—	1	—
(8) + 1	—	—	—	—	—	1	—	—	1	—
	142	142	155	155	183	183	106	150	221	221

⁷ Where the bundles were united in a ring, the number refers to number of protoxylem strands.

observed, for the zone within which the hexarch condition persists is narrow and its level is variable; and there is necessarily more or less variation in the level at which the sections are cut.

2. *Base of Hypocotyl.* In the series of sections of the base of the hypocotyl made at Storrs, the number of double vascular strands (each of which is derived from a primary root bundle and corresponds to a pole of the root) and the number of intercalary strands were recorded separately. There is no difficulty in distinguishing between these two categories of bundles, since the latter are almost invariably without protoxylem elements and are irregularly placed.

The original data for the five lines are condensed in table 2. The number of bundle pairs (the primary double bundles) is given in parenthesis, and the number of intercalary bundles, if such are present, follows the + sign outside the parenthesis.

There are three outstanding features in this table.

First, the wide range of variation in the number and in the combinations of primary double bundles and intercalary bundles in both normal and abnormal plants observed when reasonably large series of seedlings are sectioned. It is clear that an anatomist who deals with only a few seedlings may obtain an altogether inadequate picture of the conditions which actually prevail in the species under investigation.

Second, notwithstanding the wide range of variation there are conspicuous modal classes in both normal and abnormal seedlings. In the normal plants these fall in all cases on four primary double bundles, without intercalary bundles, or with but one intercalary bundle; and in the trimerous plants, on six primary double bundles without intercalary bundles.

Third, the plants which are externally dimerous and trimerous are also clearly differentiated in internal morphology. The internal characters are, however, transgressive. It is impossible in some cases to distinguish from sections of the hypocotyl base alone between plants which superficially fall into the strictly alternative classes of dimery and trimery.

For purposes of more detailed analysis these formulae must be split up into their component elements.

A. Primary Double Bundles. The distribution of the number of primary double bundles in the five lines considered is shown in table 3 for dimerous and trimerous seedlings. These frequencies, reduced to a percentage basis, are represented graphically in figure 13. This shows that in all five lines the modal number of primary double bundles is two higher in the trimerous than in the dimerous plants. In the dimerous plants the modal class is in all cases 4; in the trimerous seedlings the modal class is 6. There is, therefore, a profound reorganization in the vascular anatomy of the seedling upon the assumption of a trimerous external organization.

Limiting our attention to primary double bundles and judging from modal classes only, an increase of fifty percent in the number of vascular elements is

TABLE 3. *Number of primary double bundles at base of hypocotyl in trimerous and dimerous seedlings*

	4	5	6	7	8	Total
Line 75						
Trimerous	1	11	121	8	1	142
Percent	0.70	7.75	85.21	5.63	0.70	
Dimerous	117	19	6	—	—	142
Percent	82.39	13.38	4.23			
Line 93						
Trimerous	1	18	132	4	—	155
Percent	0.65	11.61	85.16	2.58		
Dimerous	90	50	15	—	—	155
Percent	58.06	32.26	9.68			
Line 98						
Trimerous	1	11	170	1	—	183
Percent	0.55	6.01	92.90	0.55		
Dimerous	165	16	1	—	1	183
Percent	90.16	8.74	0.55		0.55	
Line 139						
Trimerous	1	8	97	—	—	106
Percent	0.94	7.55	91.51			
Dimerous	147	3	—	—	—	150
Percent	98.00	2.00				
Line 143						
Trimerous	5	46	159	9	2	221
Percent	2.26	20.81	71.94	4.07	0.90	
Dimerous	209	10	1	1	—	221
Percent	94.57	4.52	0.45	0.45		

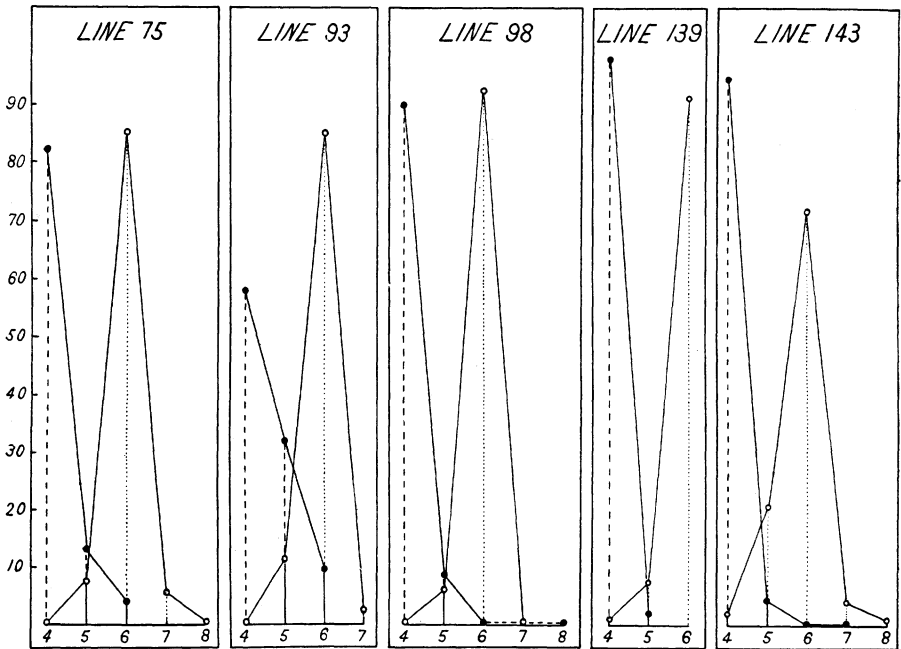


FIG. 13. Percentage frequency distribution for number of primary double bundles at base of hypocotyl in dimerous (solid dots) and trimerous (circles) seedlings.

associated with an increase of fifty percent in the number of cotyledons and leaves. The distributions show, however, that this is only an incomplete, and to some extent an erroneous, statement of the condition. In the dimerous seedlings the modal number of primary double bundles is 4, and all departures from the modal number are higher. In the trimerous seedlings the modal number is 6, and the departures may be in either the positive or the negative direction. The frequency distribution for the dimerous plants is therefore wholly skew, forming a typical J-curve; that for the trimerous plants more or less symmetrical,⁸ but with departures occurring chiefly as smaller numbers of bundles.

The variation of primary double bundle number in dimerous and trimerous plants is, therefore, transgressive. The number of externally dimerous seedlings which might be considered to be anatomically trimerous, and the number of trimerous seedlings which might on anatomical grounds be considered dimerous is, however, very small.

Turning to the physical constants in table 4, we note that the mean

TABLE 4. *Statistical constants for number of primary double bundles at base of hypocotyl of trimerous plants and their normal controls*

	Mean	Standard Deviation	Coefficient of Variation
Line 75			
Trimerous (N = 142).....	5.98 ± .02	0.436 ± .017	7.28 ± .29
Dimerous (N = 142).....	4.22 ± .03	0.505 ± .020	11.97 ± .49
Actual difference.....	+1.76 ± .04	-0.069 ± .026	-4.69 ± .56
Relative difference.....	41.71	13.66	
Line 93			
Trimerous (N = 155).....	5.90 ± .02	0.396 ± .015	6.72 ± .26
Dimerous (N = 155).....	4.52 ± .04	0.666 ± .026	14.74 ± .58
Actual difference.....	+1.38 ± .04	-0.270 ± .030	-8.02 ± .63
Relative difference.....	30.53	40.54	
Line 98			
Trimerous (N = 183).....	5.93 ± .01	0.288 ± .010	4.86 ± .17
Dimerous (N = 183).....	4.12 ± .02	0.427 ± .015	10.36 ± .37
Actual difference.....	+1.81 ± .02	-0.139 ± .018	-5.50 ± .41
Relative difference.....	43.93	32.55	
Line 139			
Trimerous (N = 106).....	5.91 ± .02	0.323 ± .015	5.47 ± .25
Dimerous (N = 150).....	4.02 ± .01	0.140 ± .005	3.48 ± .14
Actual difference.....	+1.89 ± .02	+0.183 ± .016	+1.99 ± .28
Relative difference.....	47.01	130.71	
Line 143			
Trimerous (N = 221).....	5.81 ± .03	0.581 ± .019	10.01 ± .32
Dimerous (N = 221).....	4.07 ± .01	0.315 ± .010	7.75 ± .25
Actual difference.....	+1.74 ± .03	+0.266 ± .021	+2.26 ± .41
Relative difference.....	42.75	84.44	

⁸ Line 139 is probably only an apparent exception to this rule. In both dimerous and trimerous seedlings variations from the modal class are extremely rare, and variations above the modal class have not been found in the 106 trimerous seedlings of this line sectioned.

number of primary double bundles at the base of the hypocotyl of trimerous plants is from 1.38 to 1.89 higher than in the dimerous controls. This represents an excess of from 30.5 to 47.0 percent.

The five lines are not, however, consistent in the relative variability of the normal and abnormal seedlings.

The standard deviation of the number of primary double bundles in the trimerous plants is lower than that in the dimerous plants in lines 75, 93, and 98. The differences are from 13.7 to 40.5 percent of the control values. Lines 139 and 143 are in contrast to the foregoing. The trimerous plants of line 139 have a standard deviation of $0.323 \pm .015$ bundles, whereas the dimerous controls have a standard deviation of $0.140 \pm .005$, giving a difference of $+.183 \pm .016$, which is 11.4 times as large as its probable error. In line 143 the trimerous plants have a standard deviation of $0.581 \pm .019$ bundles as compared with $0.315 \pm .010$ bundles in the normal controls, giving a difference of $+.266 \pm .021$, which is 12.7 times as large as its probable error. These are relative differences of +130.7 percent for line 139 and +84.4 percent for line 143.

The same differences in variability between the lines is also conspicuous in the relative variabilities as measured by the coefficients of variation. In the first three lines (75, 93, and 98) the coefficients of variation in the trimerous plants range from 4.9 to 7.3 percent as compared with 10.4 to 14.7 percent in the dimerous controls, giving differences in relative

TABLE 5. *Number of intercalary bundles at base of hypocotyl in trimerous and dimerous seedlings*

	0	1	2	3	4	5	6	Total
Line 75								
Trimerous.....	116	20	5	—	—	—	1	142
Percent.....	81.69	14.08	3.52				0.70	
Dimerous.....	87	35	11	5	2	2	—	142
Percent.....	61.27	24.65	7.75	3.52	1.41	1.41		
Line 93								
Trimerous.....	129	21	4	—	1	—	—	155
Percent.....	83.23	13.55	2.58		0.65			
Dimerous.....	66	58	24	6	1	—	—	155
Percent.....	42.58	37.42	15.48	3.87	0.65			
Line 98								
Trimerous.....	165	16	1	—	—	1	—	183
Percent.....	90.16	8.74	0.55			0.55		
Dimerous.....	104	52	24	3	—	—	—	183
Percent.....	56.83	28.42	13.11	1.64				
Line 139								
Trimerous.....	96	10	—	—	—	—	—	106
Percent.....	90.57	9.43						
Dimerous.....	139	11	—	—	—	—	—	150
Percent.....	92.67	7.33						
Line 143								
Trimerous.....	157	64	—	—	—	—	—	221
Percent.....	71.04	28.95						
Dimerous.....	156	61	4	—	—	—	—	221
Percent.....	70.58	27.60	1.80					

variability ranging from -4.7 to -8.0 percent. In line 139 the coefficient of variation for trimerous seedlings is 5.47, whereas that for dimerous seedlings is 3.48. In line 143, the coefficient of variation for trimerous seedlings is 10.01, whereas that for dimerous seedlings is 7.75. Thus the relative variability in these two lines is greater in the *trimerous* than in the *dimerous* seedlings.

B. Intercalary Bundles. The distribution of the number of intercalary bundles (considered alone) in the base of the hypocotyl is shown in table 5.

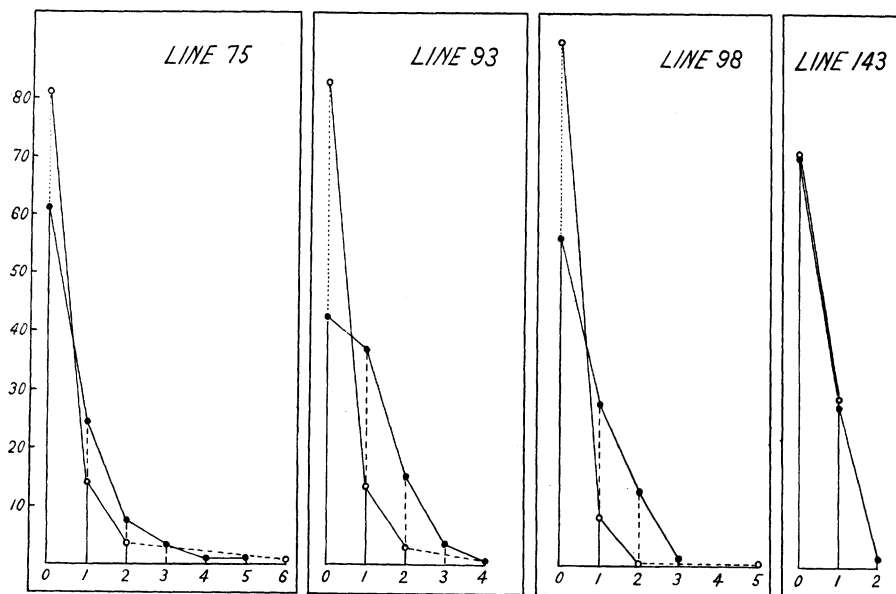


FIG. 14. Percentage frequency distribution of number of intercalary bundles at base of hypocotyl in dimerous (solid dots) and trimerous (circles) seedlings.

The graphs in figure 14 show that for both dimerous and trimerous seedlings *no* intercalary bundles is the modal condition. In both cases the distribution is wholly skew. The normal and the abnormal seedlings of lines 75, 93, and 98 differ conspicuously, however, in that the percentage of seedlings with no intercalary bundles is much higher in the trimerous seedlings, while, conversely, the percentage of seedlings with from 1 to 5 intercalary bundles is much higher in the dimerous plants. These differences are not found in lines 139 and 143. As a matter of fact, the percentage of seedlings with no intercalary bundles is slightly, but perhaps not significantly, higher in the dimerous seedlings of line 139. In both lines 139 and 143 the number of seedlings with 1 or 2 intercalary bundles is very small indeed in both trimerous and dimerous series. The two lines are essentially alike in this regard and line 143 only is represented on the diagram.

The percentages of the seedlings with no intercalary bundles in the two classes of plants and the differences in the percentage are as follows:

	Trimerous	Dimerous	Difference
Line 75.....	81.69	61.27	+20.42
Line 93.....	83.23	42.58	+40.65
Line 98.....	90.16	56.83	+33.33
Line 139.....	90.57	92.67	- 2.10
Line 143.....	71.04	70.58	+ 0.46

The physical constants in table 6 show that the mean number of intercalary bundles in both normal and abnormal seedlings is small—less than a single bundle per plant in every case.

TABLE 6. *Statistical constants for number of intercalary bundles at base of hypocotyl of trimerous plants and their normal controls*

	Mean	Standard Deviation	Coefficient of Variation
Line 75			
Trimerous (N = 142).....	.25±.04	0.686±.027	270.69±42.86
Dimerous (N = 142).....	.63±.06	1.024±.041	161.60±16.13
Actual difference.....	-.38±.07	-0.338±.049	+109.09±45.79
Relative difference.....	60.32	33.00	
Line 93			
Trimerous (N = 155).....	.21±.03	.545±.021	255.80±36.78
Dimerous (N = 155).....	.83±.05	.874±.033	105.79± 7.29
Actual difference.....	-.62±.06	-.329±.039	+150.01±37.50
Relative difference.....	74.69	37.64	
Line 98			
Trimerous (N = 183).....	.13±.02	.480±.017	381.67±73.88
Dimerous (N = 183).....	.60±.04	.776±.027	130.21± 9.62
Actual difference.....	-.47±.04	-.296±.032	+251.46±74.50
Relative difference.....	78.33	38.14	
Line 139			
Trimerous (N = 106).....	.09±.02	.292±.014	309.84±64.50
Dimerous (N = 150).....	.07±.01	.261±.010	355.48±70.95
Actual difference.....	+.02±.02	+.031±.017	- 45.64±95.89
Relative difference.....	28.57	11.88	
Line 143			
Trimerous (N = 221).....	.29±.02	.454±.015	156.62±12.21
Dimerous (N = 221).....	.31±.02	.501±.016	160.44±12.76
Actual difference.....	-.02±.03	-.047±.021	- 3.82±17.66
Relative difference.....	6.45	9.38	

Again the lines fall into two classes, those in which the number of intercalary bundles is conspicuously higher in the dimerous plants (lines 75, 93, and 98) and those in which the numbers are essentially identical (lines 139 and 143). In the trimerous seedlings of the first group the average number ranges from 0.13 to 0.25, whereas in the dimerous it varies from 0.60 to 0.83 bundle. Thus the mean number of intercalary bundles is

from 60 to 78 percent smaller in the trimerous than in the dimerous seedlings.

In line 139 the mean number of intercalary bundles is actually larger in the trimerous seedlings, but the difference is only $+.02 \pm .02$.

In line 143 the mean number of intercalary bundles in trimerous and dimerous seedlings is practically identical, the difference being only $-.02 \pm .03$. In both of these lines the differences are insignificant in comparison with their probable errors.

It is also interesting to note that in lines 75, 93, and 98 the differentiation between abnormal and normal seedlings is greater with respect to the number of intercalary bundles than with respect to primary double bundles. Turning back to table 4, we note that the number of primary double bundles is from 31 to 44 percent higher in the trimerous plants, whereas the number of intercalary bundles is from 60 to 78 percent lower. In lines 139 and 143 the difference in the mean of the number of primary double bundles of trimerous and dimerous plants is practically the same as in the other lines, but in these lines the two types of seedlings are essentially identical in number of intercalary bundles.

If we consider the comparative variability of dimerous and trimerous seedlings as to intercalary bundle number, we find that here, as in the case of number of primary double bundles, the lines differ among themselves. In all lines except 139 the standard deviations of number of intercalary bundles in the trimerous seedlings are smaller than in the dimerous. In lines 75, 93, and 98 the constants for the trimerous seedlings are from 33 to 38 percent smaller than those of the dimerous controls. In line 143 the difference has the same sign but is only -9.38 percent of the control value. In line 139 the difference is $+11.88$ percent.

The coefficients of variation are very high in both normal and abnormal seedlings, and this great variation renders the probable errors of little value as criteria of statistical significance of differences between the two types of seedlings. In lines 75, 93, and 98, the coefficients of variation for trimerous plants are conspicuously higher than those for the dimerous controls. In line 143 the coefficients of variation for the two types of seedlings are practically the same. In line 139, however, the coefficient of variation for the number of intercalary bundles is higher in dimerous than in trimerous plants.

C. Total Bundles. Having considered the frequency distribution and statistical constants for the two types of vascular structures found in the base of the hypocotyl, it is now desirable to combine the two types of bundles in order to consider the total number of vascular elements at this level.

This problem presents certain morphological difficulties. The primary double bundles are each derived from a single root pole, and do not become clearly divided into two bundles until the level of transition is reached from root structure to stem structure at the base of the hypocotyl. Many of the intercalary bundles appear at this level or a little lower. In determining

TABLE 7. *Total number of bundles at base of hypocotyl in trimerous and dimerous seedlings. Primary double bundles are counted as one bundle only*

	4	5	6	7	8	9	10	Total
Line 75								
Trimerous	—	1	115	21	3	1	1	142
Percent	—	0.70	80.99	14.79	2.11	0.70	0.70	
Dimerous	69	43	19	6	3	2	—	142
Percent	48.59	30.28	13.38	4.23	2.11	1.41	—	
Line 93								
Trimerous	—	5	130	18	2	—	—	155
Percent	—	3.23	83.87	11.61	1.29	—	—	
Dimerous	34	59	41	17	4	—	—	155
Percent	21.94	38.06	26.45	10.97	2.58	—	—	
Line 98								
Trimerous	—	4	166	12	—	1	—	183
Percent	—	2.19	90.71	6.56	—	0.55	—	
Dimerous	97	49	32	3	1	1	—	183
Percent	53.01	26.78	17.49	1.64	0.55	0.55	—	
Line 139								
Trimerous	—	5	96	5	—	—	—	106
Percent	—	4.72	90.57	4.72	—	—	—	
Dimerous	138	10	2	—	—	—	—	150
Percent	92.00	6.67	1.33	—	—	—	—	
Line 143								
Trimerous	2	18	165	30	5	1	—	221
Percent	0.90	8.14	74.66	13.57	2.26	0.45	—	
Dimerous	150	63	9	2	—	—	—	221
Percent	67.87	27.15	4.0	0.91	—	—	—	

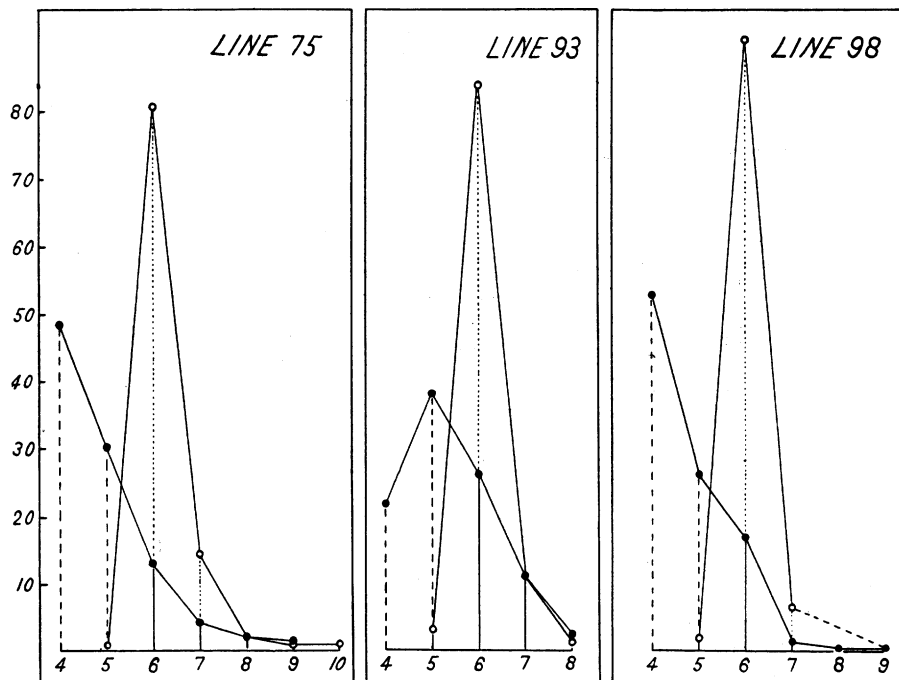


FIG. 15. Percentage frequency distribution of total bundles at base of hypocotyl. Primary double bundles counted as single bundles.

the total number of bundles at the base of the hypocotyl, it therefore becomes a question as to whether we should count each primary double bundle as a single strand or as a double strand; adding, of course, the number of intercalary bundles in each case.

The distribution of total bundle number at this level according to the former method (primary double bundles counted as one, plus intercalaries) is shown in table 7, for both dimerous and trimerous seedlings. The results are shown clearly in figure 15.⁹ The modal number is on 4 (lines 75, 98, 139, and 143) or 5 (line 93) bundles in the case of the dimerous seedlings, but invariably on 6 in the trimerous plantlets of the five lines. The distribution of number of bundles is almost wholly skew in the case of the normal seedlings, line 93 being slightly different from the others, but fairly symmetrical in the trimerous series.

The constants given in table 8 show that on the average the trimerous plants have from 0.77 to 1.91 bundles more than the dimerous plants. This is an excess of from 14.4 to 46.7 percent instead of the 50 percent which one might expect if the increase in number of bundles were proportional to the number of cotyledons or primordial leaves.

TABLE 8. *Statistical constants for total number of bundles at base of hypocotyl of trimerous plants and their normal controls. Primary double bundles are counted as one bundle only*

	Mean	Standard Deviation	Coefficient of Variation
Line 75			
Trimerous (N = 142)	6.23 ± .03	0.601 ± .024	9.65 ± .39
Dimerous (N = 142)	4.85 ± .06	1.087 ± .044	22.41 ± .94
Actual difference	+1.38 ± .07	-0.486 ± .050	-12.76 ± 1.01
Relative difference	28.45	44.71	
Line 93			
Trimerous (N = 155)	6.11 ± .02	0.434 ± .017	7.10 ± .27
Dimerous (N = 155)	5.34 ± .06	1.019 ± .039	19.07 ± .76
Actual difference	+0.77 ± .06	-0.585 ± .042	-11.97 ± .80
Relative difference	14.41	57.41	
Line 98			
Trimerous (N = 183)	6.06 ± .02	0.365 ± .013	6.02 ± .21
Dimerous (N = 183)	4.72 ± .05	0.909 ± .032	19.28 ± .70
Actual difference	+1.34 ± .05	-0.544 ± .035	-13.26 ± .73
Relative difference	28.39	59.85	
Line 139			
Trimerous (N = 106)	6.00 ± .02	0.307 ± .014	5.12 ± .24
Dimerous (N = 150)	4.09 ± .02	0.334 ± .013	8.15 ± .32
Actual difference	+1.91 ± .03	-0.027 ± .019	-3.03 ± .40
Relative difference	46.70	8.08	
Line 143			
Trimerous (N = 221)	6.10 ± .03	0.613 ± .020	10.06 ± .33
Dimerous (N = 221)	4.38 ± .03	0.609 ± .020	13.91 ± .45
Actual difference	+1.72 ± .04	+0.004 ± .028	-3.85 ± .56
Relative difference	39.27	0.66	

⁹ Lines 139 and 143 are in essential agreement with 75, 93, and 98, and are not drawn.

The variability, both absolute and relative, of the number of bundles is higher in dimerous than in trimerous plants. It is conspicuously higher in lines 75, 93, and 98. Thus the standard deviations for the trimerous plants range from 0.37 to 0.60 in the three lines as compared with 0.91 to 1.09 in the dimerous controls. The relative differences show that the variability of the trimerous plants is from 45 to 60 percent less than that of the dimerous plants. In the case of line 143, however, the difference between the standard deviation of the two types of seedlings is very small—less, indeed, than the probable error of the difference. Practically the same condition is found in line 139.

The coefficients of variation show that the trimerous plants have a variability in bundle number which is from 5.1 to 10.1 percent of the mean number of bundles, whereas the dimerous controls have a variability which is from 8.2 to 22.4 percent of the average number. In lines 75, 93, and 98 the difference between the two types is much more conspicuous than in lines 139 and 143.

Since in practically all cases, however, the primary double bundles have already clearly become two strands at the point where the intercalaries appear, it probably gives us a better conception of total bundle number here to count each primary bundle as *two*, and to add thereto the number of intercalaries. The actual and the percentage distribution according to this method are shown in table 9. Lines 75, 93, and 139 are represented in

TABLE 9. *Total number of bundles at base of hypocotyl in trimerous and dimerous seedlings. Primary double bundles are counted as two*

	8	9	10	11	12	13	14	15	16	17	Total
Line 75											
Trimerous ..	—	—	2	8	109	12	10	—	1	—	142
Percent ..			1.41	5.63	76.76	8.45	7.04		0.70		
Dimerous...	69	30	23	8	8	4	—	—	—	—	142
Percent ..	48.59	21.13	16.20	5.63	5.63	2.82					
Line 93											
Trimerous ..	—	—	5	10	124	11	5	—	—	—	155
Percent ..			3.23	6.45	80.00	7.10	3.23				
Dimerous...	34	37	35	23	20	4	2	—	—	—	155
Percent ..	21.93	23.87	22.58	14.84	12.90	2.58	1.29				
Line 98											
Trimerous ..	—	1	4	6	161	10	1	—	—	—	183
Percent ..		0.55	2.19	3.28	87.98	5.46	0.55				
Dimerous...	97	43	29	10	2	1	—	—	—	1	183
Percent ..	53.01	23.50	15.85	5.46	1.09	0.55				0.55	
Line 139											
Trimerous ..	—	1	4	4	92	5	—	—	—	—	106
Percent ..		0.94	3.77	3.77	86.79	4.72					
Dimerous...	138	9	1	2	—	—	—	—	—	—	150
Percent ..	92.00	6.00	0.67	1.33							
Line 143											
Trimerous ..	2	3	15	31	134	25	5	4	1	1	221
Percent ..	0.90	1.36	6.79	14.03	60.63	11.31	2.26	1.81	0.45	0.45	
Dimerous...	150	55	9	5	—	1	1	—	—	—	221
Percent ..	67.87	24.89	4.07	2.26		0.45	0.45				

TABLE 10. Statistical constants for total number of bundles at base of hypocotyl of trimerous plants and their normal controls. Primary double bundles are counted as two

	Mean	Standard Deviation	Coefficient of Variation
Line 75			
Trimerous (N = 142)	12.17 ± .04	0.750 ± .030	6.17 ± .25
Dimerous (N = 142)	9.07 ± .08	1.351 ± .054	14.90 ± .61
Actual difference	+3.10 ± .08	−0.601 ± .062	−8.73 ± .66
Relative difference	34.18	44.49	
Line 93			
Trimerous (N = 155)	12.01 ± .03	0.627 ± .024	5.22 ± .20
Dimerous (N = 155)	9.86 ± .08	1.483 ± .057	15.04 ± .59
Actual difference	+2.15 ± .08	−0.856 ± .062	−9.82 ± .62
Relative difference	21.81	57.72	
Line 98			
Trimerous (N = 183)	11.97 ± .02	0.495 ± .018	4.14 ± .15
Dimerous (N = 183)	8.84 ± .06	1.190 ± .042	13.47 ± .48
Actual difference	+3.13 ± .06	−0.695 ± .046	−9.33 ± .50
Relative difference	35.41	58.40	
Line 139			
Trimerous (N = 106)	11.91 ± .04	0.558 ± .026	4.69 ± .22
Dimerous (N = 150)	8.11 ± .02	0.440 ± .017	5.43 ± .21
Actual difference	+3.80 ± .04	+0.118 ± .031	−0.74 ± .30
Relative difference	46.85	26.82	
Line 143			
Trimerous (N = 221)	11.90 ± .05	1.105 ± .035	9.28 ± .30
Dimerous (N = 221)	8.45 ± .04	.831 ± .027	9.84 ± .32
Actual difference	+3.45 ± .06	+0.274 ± .044	−0.56 ± .44
Relative difference	40.83	32.97	

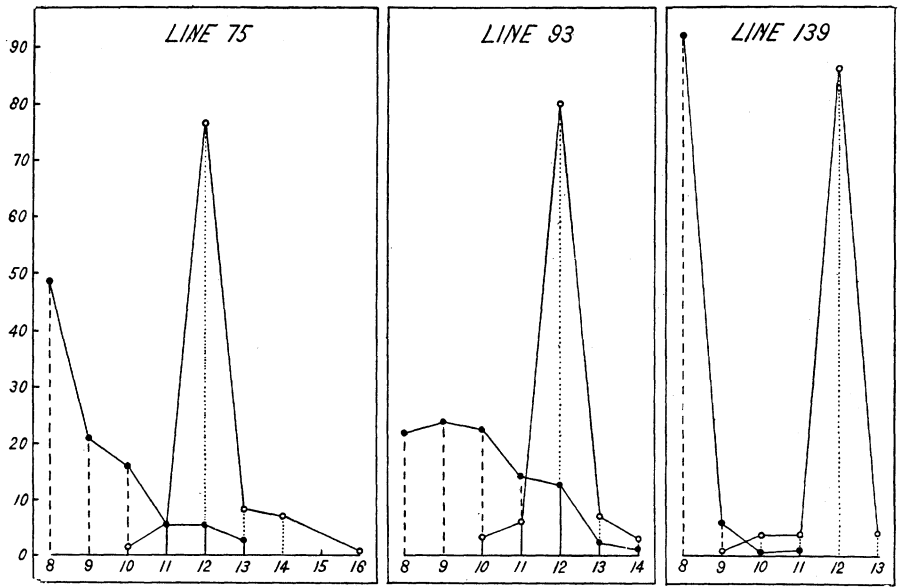


FIG. 16. Percentage frequency distribution of total bundles at base of hypocotyl in dimerous and trimerous seedlings. Primary double bundles counted as two.

figure 16. Comparison of these figures with those in figure 15 shows essentially the same type of distribution for the dimerous and trimerous plants. The grades of the classes are merely about double what they were in the former method of treatment.

The statistical constants are compared in table 10.

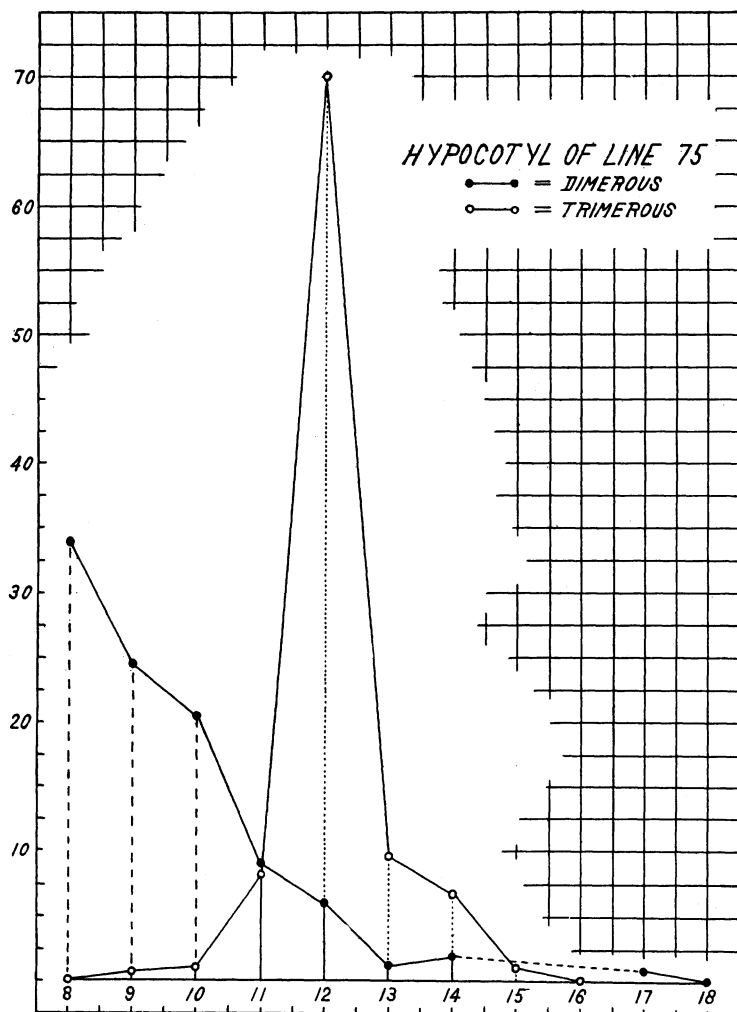


FIG. 17. Percentage frequency distribution of total bundles in central region of hypocotyl.

For all five lines the constants show a higher mean number of bundles in the trimerous than in the dimerous seedlings, the mean being approximately 12 in the former and 8 or 10 in the latter. Thus the trimerous seedlings have from 21.8 to 46.9 percent more bundles than the dimerous seedlings.

TABLE II. *Number of bundles in central region of hypocotyl of trimerous and dimerous seedlings*

	8	9	10	11	12	13	14	15	16	17	18	19	20	Total
Line 75														
Trimerous.....	1	3	5	36	292	40	29	5	1	4	—	—	—	416
Percent.....	0.24	0.72	1.20	8.65	70.19	9.62	6.97	1.20	0.24	0.96	—	—	—	
Dimerous.....	143	103	86	38	26	7	9	—	—	3	1	—	—	416
Percent.....	34.37	24.76	20.67	9.13	6.25	1.68	2.16	—	—	0.72	0.24	—	—	
Line 93.....														
Trimerous.....	—	—	8	32	382	82	38	12	1	—	1	—	1	557
Percent.....	—	—	1.44	5.75	68.58	14.72	6.82	2.15	0.18	—	0.18	—	0.18	
Dimerous.....	34	93	169	105	96	39	18	1	—	—	2	—	—	557
Percent.....	6.10	16.70	30.34	18.85	17.24	7.00	3.23	0.18	—	—	0.36	—	—	
Line 98.....														
Trimerous.....	—	1	6	12	297	21	8	—	—	—	—	—	—	345
Percent.....	—	0.29	1.74	3.48	86.09	6.09	2.32	—	—	—	—	—	—	
Dimerous.....	113	110	77	32	9	3	—	—	—	1	—	—	—	345
Percent.....	32.75	31.88	22.32	9.28	2.61	0.87	—	—	—	0.29	—	—	—	
Line 139.....														
Trimerous.....	—	—	4	8	84	6	3	1	—	—	—	—	—	106
Percent.....	—	—	3.77	7.55	79.25	5.66	2.83	0.94	—	—	—	—	—	
Dimerous.....	137	10	2	1	—	—	—	—	—	—	—	—	—	150
Percent.....	91.33	6.67	1.33	0.67	—	—	—	—	—	—	—	—	—	
Line 143.....														
Trimerous.....	2	1	11	14	136	21	25	6	3	1	1	—	—	221
Percent.....	0.90	0.45	4.98	6.33	61.54	9.50	11.31	2.71	1.36	0.45	0.45	—	—	
Dimerous.....	138	41	25	10	2	3	1	1	—	—	—	—	—	221
Percent.....	62.44	18.55	11.31	4.52	0.90	1.36	0.45	0.45	—	—	—	—	—	

In variability as measured by coefficient of variation, the dimerous plants exceed the trimerous throughout, conspicuously so in lines 75, 93, and 98. In their standard deviation, the dimerous also markedly exceed the trimerous in these three lines, but in lines 139 and 143 the trimerous plants slightly exceed the dimerous.

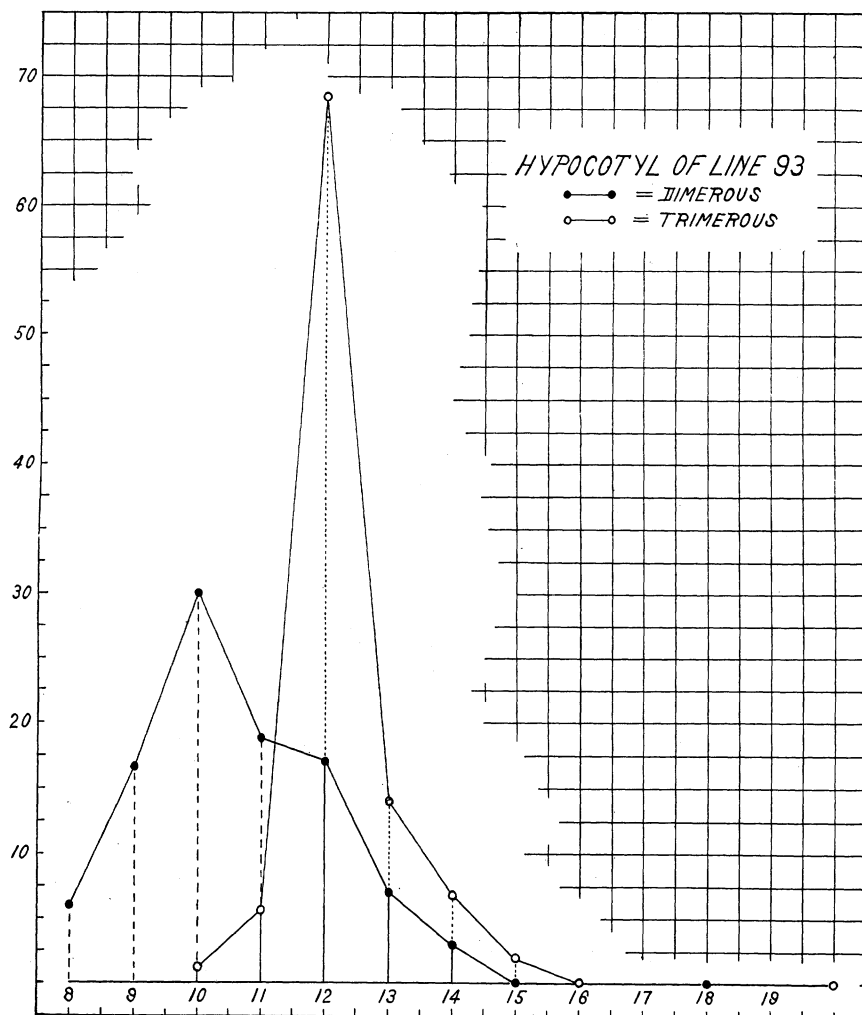


FIG. 18. Percentage frequency distribution of total bundles in central region of hypocotyl.

D. Summary for Base of Hypocotyl. For the base of the hypocotyl, therefore, it is evident that in total bundle number the trimerous seedlings decidedly exceed the dimerous ones. The intercalary bundles alone (which form but a small part of the total) tend to be more numerous in the dimerous seedlings.

In variability in bundle number at this region, dimerous seedlings in general exceed trimerous ones; although two of the five lines studied furnish slight exceptions to this rule.

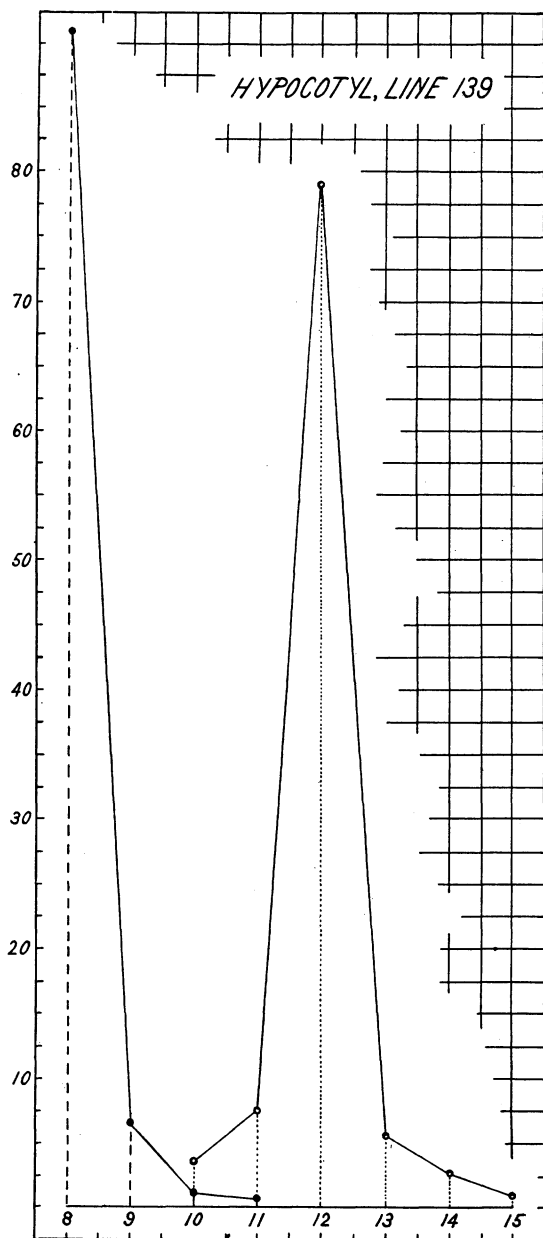


FIG. 19. Percentage frequency distribution of total bundles in central region of hypocotyl.

3. *Central Region of Hypocotyl.* In the sections made in the central regions of the hypocotyl and of the epicotyl at both Cold Spring Harbor and Storrs, the total number of bundles was counted, no distinction being made between the bundles originating from the primary double bundles and those of intercalary origin.

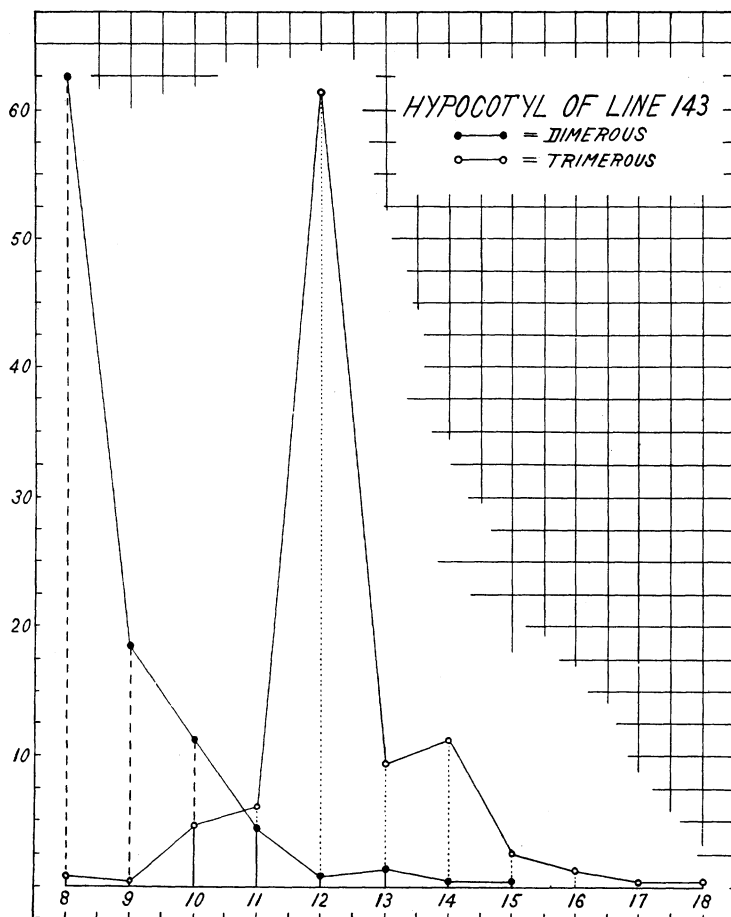


FIG. 20. Percentage frequency distribution of total bundles in central region of hypocotyl.

The frequency distributions are shown in table 11. The relative frequencies for line 75 are shown in figure 17. The form of the distributions in line 98 is in essential agreement with those in line 75 and is not represented. The distributions for line 93 are represented in figure 18. The distribution for line 139 is shown in figure 19. That for line 143 appears in figure 20.

The conspicuous feature of these distributions is the wide variation in bundle number and the conspicuous skewness of the frequencies for the

normal plants of lines 75, 93, and 98. In these, bundle number ranges from 8 to 18 with a relatively large number of bundles in the lower classes. The modal number of bundles in the hypocotyl of normal seedlings of lines 75, 98, 139, and 143 is 8, while in line 93 it is 10.

The normal plants of the five lines differ conspicuously in variability. The number of seedlings falling in the modal class is relatively small and the range of variation relatively wide in lines 75, 93, and 98 as compared with line 139. Line 143 occupies an intermediate position in this regard.

In all the lines except 93 the distribution of number of bundles in the hypocotyl of normal seedlings is wholly skew, the frequency decreasing from the modal number (eight) towards the upper end of the range. In line 93 (figure 18) the distribution is also skew, but the frequency decreases from the modal number (ten) towards both ends of the range.

In the trimerous plantlets of all five series the modal number of bundles in the mid-region of the hypocotyl is 12. The extent of concentration into the modal class and the range of variation differs greatly in the five lines. This is very narrow in lines 98 and 139 and relatively wide in line 143.

The frequency distribution and figures bring out very clearly indeed the differentiation of the trimerous and dimerous seedlings in the number of vascular bundles.

TABLE 12. *Statistical constants for number of bundles in hypocotyl of trimerous and dimerous seedlings*

	Mean	Standard Deviation	Coefficient of Variation
Line 75			
Trimerous (N = 416)	12.19 ± .03	0.982 ± .023	8.06 ± .19
Dimerous (N = 416)	9.49 ± .05	1.645 ± .039	17.34 ± .42
Actual difference	+2.70 ± .06	-0.663 ± .045	-9.28 ± .46
Relative difference	28.45	40.30	
Line 93			
Trimerous (N = 557)	12.29 ± .03	0.922 ± .019	7.50 ± .15
Dimerous (N = 557)	10.62 ± .04	1.525 ± .031	14.36 ± .30
Actual difference	+1.67 ± .05	-0.603 ± .036	-6.86 ± .34
Relative difference	15.73	39.54	
Line 98			
Trimerous (N = 345)	12.03 ± .02	0.532 ± .014	4.42 ± .11
Dimerous (N = 345)	9.22 ± .04	1.197 ± .031	12.99 ± .34
Actual difference	+2.81 ± .04	-0.665 ± .034	-8.57 ± .36
Relative difference	30.47	55.56	
Line 139			
Trimerous (N = 106)	11.99 ± .05	0.694 ± .032	5.78 ± .27
Dimerous (N = 150)	8.11 ± .02	0.409 ± .016	5.04 ± .20
Actual difference	+3.88 ± .05	+0.285 ± .036	+0.74 ± .34
Relative difference	47.84	69.68	
Line 143			
Trimerous (N = 221)	12.29 ± .06	1.283 ± .041	10.44 ± .34
Dimerous (N = 221)	8.71 ± .05	1.187 ± .038	13.63 ± .45
Actual difference	+3.58 ± .08	+0.096 ± .056	-3.19 ± .57
Relative difference	41.10	8.09	

The differences between the lines can best be seen from the figures.

For a more critical comparison we must have recourse to statistical constants and their probable errors.

The results for the hypocotyl of trimerous seedlings and their normal controls are set forth in table 12. Without exception the number of bundles in abnormal plants is higher than that in the control plants. The differences range from 1.7 to 3.9 bundles. These differences are many times as large as their probable errors and are unquestionably significant. The relative differences are about 16 percent in line 93, 30 percent in lines 75 and 98, 41 percent in line 143, and 48 percent in line 139.

Both the standard deviation and the coefficient of variation of the number of bundles in the hypocotyl are lower in the abnormal than in the normal plants in lines 75, 93, and 98. In lines 139 and 143 the relationship of the standard deviations of the trimerous and dimerous plants is exactly reversed, that of the trimerous plants being somewhat larger than that of the dimerous series. The difference in line 143 is $+.096 \pm .056$, which is nearly twice as large as its probable error and possibly statistically significant. In line 139 the difference in standard deviation is $+.285 \pm .036$. This difference is about 8 times as large as its probable error and unquestionably significant. The percentage differences in the standard deviations in lines 75, 93, and 98 range from -40 to -56 percent. In line 143 the percentage difference is $+8$ percent, while in line 139 it is $+70$ percent.

In line 143 the coefficient of variation is higher in dimerous plants (as it is in lines 75, 93, and 98), but in line 139 the trimerous show a slightly but perhaps not significantly higher relative variability.

The results as a whole show that the difference in the variability of bundle number in the two types of seedlings in lines 139 and 143 is not the same as that in lines 75, 93, and 98.

In interpreting these results we must remember that each primary double bundle at the base of the hypocotyl almost invariably divides to form two bundles at higher levels in the hypocotyl. Occasionally one of these branches may further divide into two. It is impossible in sections made in the central region of the hypocotyl to distinguish with certainty in every case between bundles originating through a division of the original protoxylem strands and those of intercalary origin.

The simplest working assumption is that the number of bundles in the central region of the hypocotyl will be given by twice the number of primary double bundles demonstrated at the base of the hypocotyl plus the number of intercalary bundles found at the base of the hypocotyl; or the number of bundles, b , at the central region should be given by

$$b = 2p + i$$

where p = primary double bundles and i = intercalary bundles.

A comparison of the number of bundles calculated by this formula with the number actually observed in the central region of the hypocotyl may be best made in a table of double entry. Table 13 gives the results for dimerous and table 14 the results for trimerous plants of line 93. The

TABLE 13. *Comparison of actual and theoretical number of bundles in hypocotyl of dimerous seedling*

Actual Number	8	9	10	11	12	13	14	Totals
Theoretical, $2p + i$ 8	12	13	6	3	—	—	—	34
9	—	14	17	3	1	1	1	37
10	—	1	22	6	5	1	—	35
11	—	—	1	9	9	3	1	23
12	—	—	—	1	14	4	1	20
13	—	—	—	—	—	1	3	4
14	—	—	—	—	—	—	2	2
Totals . .	12	28	46	22	29	10	8	155

TABLE 14. *Comparison of actual and theoretical number of bundles in hypocotyl of trimerous seedling*

Actual Number	10	11	12	13	14	15	20	Totals
Theoretical, $2p + i$ 10	1	1	3	—	—	—	—	5
11	—	6	3	—	1	—	—	10
12	—	2	102	12	6	1	1	124
13	—	—	—	8	3	—	—	11
14	—	—	—	—	4	1	—	5
Totals . .	1	9	108	20	14	2	1	155

frequencies for the cases in which the number of bundles at the mid-region of the hypocotyl calculated from the formula agrees with the number actually observed are printed in blackface type. The other lines give roughly comparable results.

It is clear that the number of hypocotyledonary bundles is not far from twice the number of primary root bundles plus the intercalary bundles. In rare cases the number of bundles in the hypocotyl is less than twice the root strands plus the number of intercalary bundles, since one of the root strands sometimes fails to divide. It may be, and not infrequently is, higher because of the appearance of extra intercalary bundles at a level higher than that sectioned at the base of the hypocotyl. In many cases the full complement of intercalary bundles has not appeared at this low level. In some cases it may be higher because of the secondary bifurcation above mentioned.

It is worth while to give the percentage frequencies of cases in which the number of bundles of the central region of the hypocotyl is given by the formula, and for comparison the relative number of cases in which it is in defect and in excess. The percentages are calculated from double entry tables like 13 and 14.

Trimerous Seedlings

	N	In Defect	$2p + i$	In Excess
Line 75.....	142	7.0	76.1	16.9
Line 93.....	155	1.3	78.1	20.7
Line 98.....	183	3.3	86.3	10.4
Line 139.....	106	7.6	80.2	12.3
Line 143.....	221	0.9	74.7	24.4

Dimerous Seedlings

	N	In Defect	$2p + i$	In Excess
Line 75.....	142	2.1	51.4	46.5
Line 93.....	155	1.9	47.7	50.3
Line 98.....	183	3.8	59.0	37.2
Line 139.....	150	0.7	98.7	0.7
Line 143.....	221	0.9	81.0	18.1

With the exception of the dimerous seedlings of line 139, the actually observed number of bundles is in excess of the number given by the formula.

In lines 75, 93, and 98 the excess is far greater in dimerous than in trimerous seedlings. Thus in the dimerous class about 40 percent of the seedlings show a number of bundles in the central region of the hypocotyl which is in excess of twice the number of primary double bundles plus the number of intercalary bundles at the base of the hypocotyl. In the case of the trimerous seedlings the excess is much smaller, being roughly 20 percent. Thus it is clear that, *especially in the normal seedlings*, a large number of the intercalary bundles do not extend to the base but appear in the axis, ending blindly below, or that a considerable proportion of the primary double bundles divide into more than two bundles.

In line 143 the number of cases in which the observed number of bundles is greater than the calculated number is much more nearly equal in the two types of seedlings. Thus in the trimerous seedlings 24.4 percent of the seedlings have a number of bundles in the central region of the hypocotyl greater than $2p + i$, whereas in the dimerous seedlings there are 18.1 percent of seedlings of this class. In line 139 only 0.7 percent of the dimerous seedlings show a number of bundles in excess of $2p + i$, whereas in the trimerous seedlings 12.3 percent are in excess.

Thus lines 139 and 143 give results diametrically opposed to those of the first three discussed.¹⁰

Summary for Central Region of Hypocotyl. It is evident from the above statements that the number of bundles in the hypocotyl of trimerous is decidedly higher than in that of dimerous seedlings; that in general the bundle number is more variable in dimerous than in trimerous seedlings; and that the intercalary bundles generally extend to a lower level in the hypocotyl of trimerous than in that of dimerous seedlings.

¹⁰ Note that the extremely small excess in line 139 may be due to the extraordinarily normal character of the vascular system of the dimerous plants of this line.

TABLE 15. *Number of bundles in central region of epicotyl of trimerous and dimerous seedlings*

	10	11	12	13	14	15	16	17	18	19	20	21	22	Total
Line 75														
Trimerous.....	—	—	3	16	63	164	93	41	27	4	4	1	—	416
Percent.....	—	—	0.72	3.85	15.14	39.42	22.36	9.86	6.49	0.96	0.96	0.24	—	416
Dimerous.....	1	4	336	46	16	10	3	—	—	—	—	—	—	416
Percent.....	0.24	0.96	80.77	11.06	3.85	2.40	0.72	—	—	—	—	—	—	416
Line 93														
Trimerous.....	—	—	5	18	47	236	129	56	51	10	4	—	1	557
Percent.....	—	—	0.90	3.23	8.44	42.37	23.16	10.05	9.16	1.80	0.72	—	0.18	557
Dimerous.....	1	6	479	42	18	10	1	—	—	—	—	—	—	557
Percent.....	0.18	1.08	86.00	7.54	3.23	1.80	0.18	—	—	—	—	—	—	557
Line 98														
Trimerous.....	—	—	8	24	69	176	49	9	7	1	1	1	—	345
Percent.....	—	—	2.32	6.96	20.00	51.01	14.20	2.61	2.03	0.29	0.29	0.29	—	345
Dimerous.....	—	—	316	23	4	1	1	—	—	—	—	—	—	345
Percent.....	—	—	91.59	6.67	1.16	0.29	0.29	—	—	—	—	—	—	345
Line 139														
Trimerous.....	—	—	—	8	21	38	24	9	4	2	—	—	—	106
Percent.....	—	—	—	7.55	19.81	35.85	22.64	8.49	3.77	1.89	—	—	—	106
Dimerous.....	—	—	131	16	3	—	—	—	—	—	—	—	—	150
Percent.....	—	—	87.33	10.67	2.00	—	—	—	—	—	—	—	—	150
Line 143														
Trimerous.....	—	—	5	9	19	54	49	37	31	9	6	2	—	221
Percent.....	—	—	2.26	4.07	8.60	24.43	22.17	16.74	14.03	4.07	2.71	0.90	—	221
Dimerous.....	—	—	169	34	11	5	2	—	—	—	—	—	—	221
Percent.....	—	—	76.47	15.38	4.98	2.26	0.90	—	—	—	—	—	—	221

4. *Central Region of Epicotyl.* The frequency distributions of the number of bundles occurring in the mid-region of the epicotyl appear in table 15 for both the abnormal and the control plants. These distributions

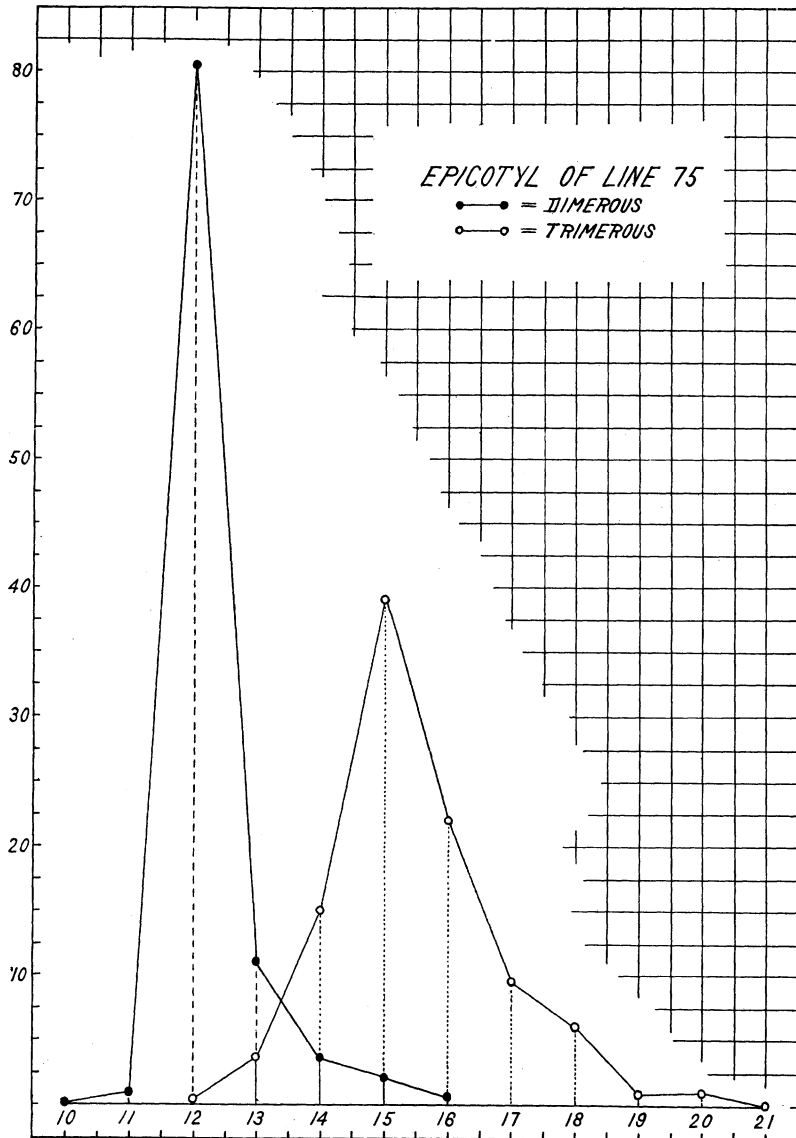


FIG. 21. Percentage frequency distribution of number of bundles in central region of epicotyl.

reduced to a percentage basis are represented graphically in figure 21 for line 75, in figure 22 for line 98, and in figure 23 for line 143. The distributions

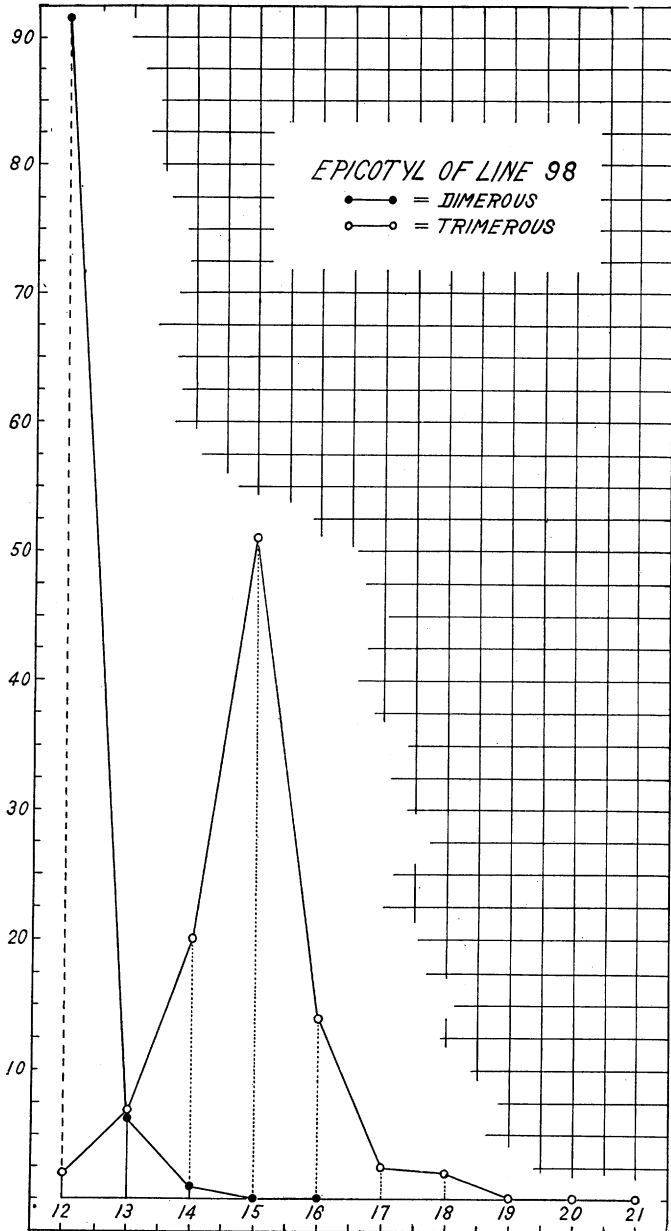


FIG. 22. Percentage frequency distribution of number of bundles in central region of epicotyl.

for line 93 are essentially the same as those for line 75. The graph for line 139 is in essential agreement with that for line 98 and is not drawn.

In the dimerous plants the difference between the form of the frequency distributions for number of epicotyledonary bundles in lines 75 and 93 on

the one hand and lines 98, 139, and 143 on the other is more apparent than real. All five lines agree in showing the frequencies for the dimerous plants largely concentrated in a single modal class with a slight but evident skew-

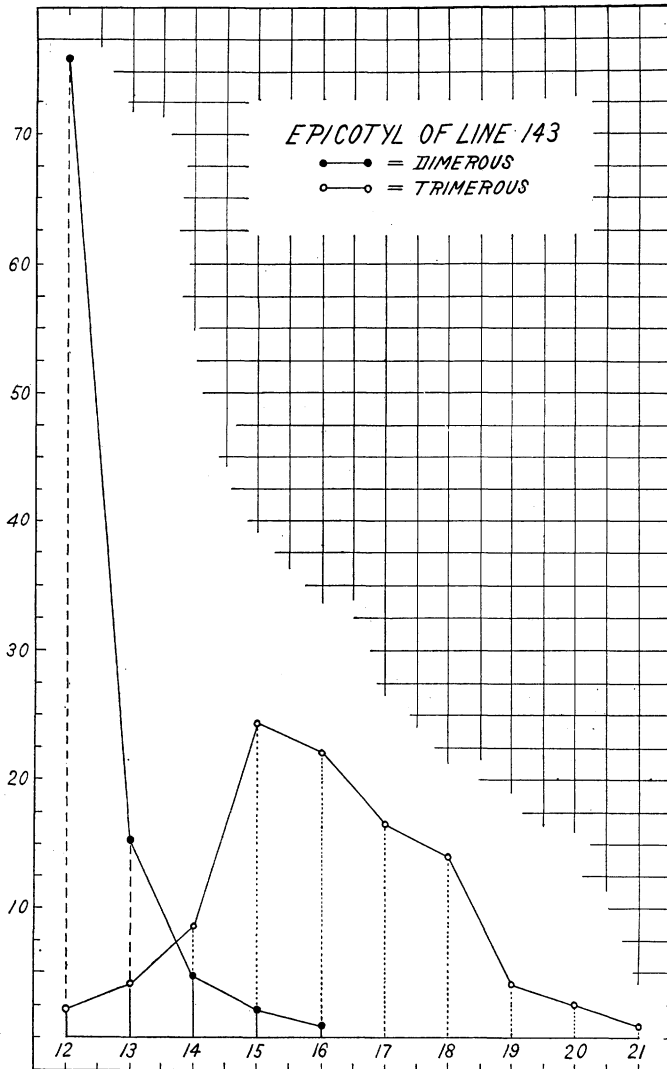


FIG. 23. Percentage frequency distribution of number of bundles in central region of epicotyl.

ness toward higher numbers of bundles. In the case of lines 75 and 93 there is a little over 1 percent of plants with fewer than the modal number of bundles, whereas in lines 98, 139, and 143 these do not occur in series of the numbers sectioned. It is quite possible that the examination of a

larger series of plantlets would result in the finding of such seedlings in lines 98, 139, and 143, thus bringing the five series into full agreement.

In the trimerous seedlings the number of bundles shows rather wide, and fairly symmetrical, distribution about the modal class, which is 15 bundles. The lines differ, however, to a considerable extent in the amount of variation from the modal class. In lines 75, 93, and 98 the frequencies are to a far greater extent concentrated into the modal class, which contains from 39 to 51 percent of the frequencies, than in line 143, which contains only 24 percent of the cases. Line 139 is intermediate between these two extremes.

For a more precise comparison we utilize the constants set forth in table 16.

TABLE 16. *Statistical constants for number of bundles in epicotyl of trimerous and dimerous seedlings*

	Mean	Standard Deviation	Coefficient of Variation
Line 75			
Trimerous (N = 416)	15.47 ± .04	1.355 ± .032	8.76 ± .21
Dimerous (N = 416)	12.27 ± .02	0.735 ± .017	5.99 ± .14
Actual difference	+3.20 ± .04	+0.620 ± .036	+2.77 ± .24
Relative difference	26.08	84.35	
Line 93			
Trimerous (N = 557)	15.65 ± .04	1.372 ± .028	8.77 ± .18
Dimerous (N = 557)	12.19 ± .02	0.615 ± .012	5.05 ± .10
Actual difference	+3.46 ± .04	+0.757 ± .030	+3.72 ± .20
Relative difference	28.38	123.09	
Line 98			
Trimerous (N = 345)	14.89 ± .04	1.152 ± .030	7.74 ± .20
Dimerous (N = 345)	12.11 ± .02	0.416 ± .011	3.44 ± .09
Actual difference	+2.78 ± .04	+0.736 ± .032	+4.30 ± .22
Relative difference	22.96	176.92	
Line 139			
Trimerous (N = 106)	15.24 ± .08	1.285 ± .060	8.44 ± .39
Dimerous (N = 150)	12.15 ± .02	0.406 ± .016	3.35 ± .13
Actual difference	+3.09 ± .08	+0.879 ± .062	+5.09 ± .41
Relative difference	25.43	216.50	
Line 143			
Trimerous (N = 221)	16.10 ± .08	1.750 ± .056	10.87 ± .35
Dimerous (N = 221)	12.36 ± .03	0.757 ± .024	6.13 ± .20
Actual difference	+3.74 ± .09	+0.993 ± .061	+4.74 ± .40
Relative difference	30.26	131.18	

These results show that without exception the average number of bundles in the epicotyl is higher in trimerous than in dimerous seedlings. The difference ranges from 2.8 to 3.7 bundles. The probable errors of these differences are so small that there can be no reasonable doubt of their significance. In relative terms, the number of bundles in the abnormal plant is from 23.0 to 30.3 percent higher than that in the normal plant.

The variability of bundle number, both absolute and relative, is far higher in the abnormal (trimerous) plants. The relative differences show that the trimerous plants are from 84 to 217 percent more variable than the dimerous in the number of bundles in the central region of the epicotyl.

We now have to consider the relative number of bundles in the hypocotyl and in the epicotyl of the same plant. The constants for the normal plants are shown in table 17 and for the trimerous seedlings in table 18.

TABLE 17. *Comparison of statistical constants for number of bundles in hypocotyl and epicotyl of same plant. Seedlings with two cotyledons and two primordial leaves*

	Mean	Standard Deviation	Coefficient of Variation
Line 75 (N = 416)			
Hypocotyl.....	9.49 ± .05	1.645 ± .039	17.34 ± .42
Epicotyl.....	12.27 ± .02	0.735 ± .017	5.99 ± .14
Actual difference.....	+2.78 ± .05	-0.910 ± .043	-11.35 ± .44
Relative difference.....	29.29	55.31	
Line 93 (N = 557)			
Hypocotyl.....	10.62 ± .04	1.525 ± .031	14.36 ± .30
Epicotyl.....	12.19 ± .02	0.615 ± .012	5.05 ± .10
Actual difference.....	+1.57 ± .04	-0.910 ± .033	-9.31 ± .32
Relative difference.....	14.78	59.67	
Line 98 (N = 345)			
Hypocotyl.....	9.22 ± .04	1.197 ± .031	12.99 ± .34
Epicotyl.....	12.11 ± .02	0.416 ± .011	3.44 ± .09
Actual difference.....	+2.89 ± .04	-0.781 ± .033	-9.55 ± .35
Relative difference.....	31.34	65.24	
Line 139 (N = 150)			
Hypocotyl.....	8.11 ± .02	0.409 ± .016	5.04 ± .20
Epicotyl.....	12.15 ± .02	0.406 ± .016	3.35 ± .13
Actual difference.....	+4.04 ± .03	-0.003 ± .023	-1.69 ± .24
Relative difference.....	49.82	0.73	
Line 143 (N = 221)			
Hypocotyl.....	8.71 ± .05	1.187 ± .038	13.63 ± .45
Epicotyl.....	12.36 ± .03	0.757 ± .024	6.13 ± .20
Actual difference.....	+3.65 ± .06	-0.430 ± .045	-7.50 ± .49
Relative difference.....	41.91	36.23	

Normal and abnormal plants have in common a larger number of bundles in the epicotyl. The differences between the means for the two organs are clearly significant in comparison with their probable errors. The percentage differences show that the epicotyl has from 15 to 50 percent more bundles than the hypocotyl.

In the dimerous seedlings the variabilities, both absolute and relative, as measured by the standard deviation and coefficient of variation, are consistent in indicating a higher variability of bundle number in the hypocotyl. The difference is, however, very slight in line 139.

The difference between the variability of the hypocotyl and that of the epicotyl in the normal seedling as measured in terms of the standard devia-

tion is from 0.8 to 0.9 bundle, or from 55 to 65 percent of the larger value in lines 75, 93, and 98. In line 143 the difference is only 0.4 bundle, or 36 percent. In line 139 there is practically no difference in the standard deviation of bundle number in the mid-region of the first two internodes of the seedling.

TABLE 18. *Comparison of statistical constants for number of bundles in hypocotyl and epicotyl of same plant. Seedlings with three cotyledons and three primordial leaves*

	Mean	Standard Deviation	Coefficient of Variation
Line 75 (N = 416)			
Hypocotyl.....	12.19±.03	0.982±.023	8.06±.19
Epicotyl.....	15.47±.04	1.355±.032	8.76±.21
Actual difference.....	+3.28±.05	+0.373±.040	+0.70±.28
Relative difference.....	26.90	37.98	
Line 93 (N = 557)			
Hypocotyl.....	12.29±.03	0.922±.019	7.50±.15
Epicotyl.....	15.65±.04	1.372±.028	8.77±.18
Actual difference.....	+3.36±.05	+0.450±.033	+1.27±.22
Relative difference.....	27.33	48.81	
Line 98 (N = 345)			
Hypocotyl.....	12.03±.02	0.532±.014	4.42±.11
Epicotyl.....	14.89±.04	1.152±.030	7.74±.20
Actual difference.....	+2.86±.04	+0.620±.033	+3.32±.22
Relative difference.....	23.77	116.54	
Line 139 (N = 106)			
Hypocotyl.....	11.99±.05	0.694±.032	5.78±.27
Epicotyl.....	15.24±.08	1.285±.060	8.44±.39
Actual difference.....	+3.25±.09	+0.591±.068	+2.66±.48
Relative difference.....	27.11	85.16	
Line 143 (N = 221)			
Hypocotyl.....	12.29±.06	1.283±.041	10.44±.34
Epicotyl.....	16.10±.08	1.750±.056	10.87±.35
Actual difference.....	+3.81±.10	+0.467±.069	+0.43±.49
Relative difference.....	31.00	36.40	

Basing the comparisons on the coefficient of variation, we note that the coefficients for the hypocotyl range from 13.0 to 17.3 percent, whereas those for the epicotyl range from 3.4 to 6.0 percent in lines 75, 93, and 98. Thus there is a difference of about 10 percent in the coefficient of variation of bundle number in the hypocotyl and epicotyl (of the normal seedlings) of these lines. In line 143 this difference is only -7.50 percent. In line 139 it is only -1.69 percent.

The statistical relationship is in full accord with the anatomical findings recorded above (p. 68) where it was shown that the intercalary bundles of the hypocotyl as they approach the cotyledonary node fuse with the (normally 8) bundles originating by the division of the (normally 4) protoxylem poles of the primary root and completely lose their individuality, exactly six bundles emerging from the complex irrespective of the number which

have entered it from the hypocotyl.¹¹ Immediately above the cotyledons the six remaining bundles approach, closing the cotyledonary gaps and forming a ring, the six members of which almost immediately divide, giving rise to the modal number, 12, which persists throughout the length of the epicotyl. It is apparently the disappearance of the intercalary bundles as a conspicuous feature of the topography which results in the lowered variability of bundle number in the epicotyl as compared with the hypocotyl.

If this conclusion be true, we should find the least difference in the variability of number of bundles in the central regions of the first two internodes in the lines in which intercalary bundles are least conspicuous as a feature of the vascular topography. As a matter of fact, this condition is strongly supported by the results for the five lines investigated. Turning back to table 6, showing the constants for number of intercalary bundles, we note that lines 75, 93, and 98 have on the average from 0.60 to 0.83 intercalary bundle per (normal) plant. These are the lines showing a relative difference of 55 to 65 percent in the standard deviations as compared with 36 percent in line 143 with an average of 0.31 intercalary bundle, and of only 0.73 percent for line 139 which has an average of only 0.07 intercalary bundle per plant. The differences in the coefficients of variation for hypocotyl and epicotyl are from -9.3 to -11.4 percent in the three lines with from 0.6 to 0.8 intercalary bundle per plant, -7.5 percent in line 143 with an average of 0.31 intercalary bundle, and only -1.7 percent in line 139 with an average of only 0.07 intercalary bundle.

In the trimerous seedlings the relationship between the variation of the number of bundles in the hypocotyl and in the epicotyl is *just the reverse* of that found in the normal type. Variability as measured by the standard deviation is significantly higher in the epicotyl of all lines studied. The same is true if the coefficient of variation be used as a measure of variability, although the differences for lines 75 and 143 are not large.

The anatomical explanation of this fact seems to be found in the peculiarities of behavior at the cotyledonary node. As pointed out above (p. 70), the epicotyledonary ring is typically made up of nine strands instead of the six characteristic of the normal plant. There is, therefore, in the modal case an increase of fifty percent in the number of bundles in the epicotyledonary ring of the trimerous plant as compared with the dimerous plant. Many of these bundles, but not all, divide to form the bundle system characteristic of the main course of the epicotyl. It is this variability in the extent of division of the bundles of the epicotyledonary ring which, in connection with the low variability of the hypocotyl due to the formation of but few intercalary bundles (except in lines 139 and 143, where the number is about the same in normal and abnormal seedlings), brings about the great variability in the bundle number of the mid-region

¹¹ This statement is based on a more detailed anatomical study of a portion of the seedlings.

of the epicotyl as compared with the mid-region of the hypocotyl, in the trimerous plants.

This condition furnishes an excellent example of the importance of a knowledge of descriptive morphology as an aid in interpreting biometric constants.

COMPARISON OF BUNDLE NUMBER IN THE FIVE LINES STUDIED

From the genetic standpoint it seems a matter of considerable interest to determine whether the three nominally pure lines¹² are differentiated with respect to their vascular anatomy.

A comparison of the percentage frequency distributions and the figures of the foregoing discussion will convince the reader that certain of the lines may be differentiated either in mean number of bundles, or in variability of number of bundles, or in both average number and variability of bundle number.

Since we hope to return to this problem later with even more extensive data, it seems unnecessary to consider the differences in the distributions and constants in detail at this time.

The results of this brief and superficial comparison seem to indicate that while different lines may not differ greatly in respect to certain of their vascular characters they may be differentiated with respect to others.

SUMMARY

This paper presents the results of a comparative and biometric study of the gross vascular anatomy of the seedling of *Phaseolus vulgaris*.

Two morphological types are considered: the normal, or dimerous, seedling with two cotyledons and two primordial leaves, and the trimerous seedling with three cotyledons and three primordial leaves.

In normal seedlings, the vascular system of the root is typically tetrarch (with four protoxylem poles), and gives rise in the base of the hypocotyl to eight bundles which continue to the cotyledonary node. From the vascular complex at this point two strands are given off to each cotyledon and six are left, each of which divides into two to produce the typical twelve-bundled condition of the epicotyl.

The trimerous seedlings typically possess six root poles instead of four, twelve bundles in the hypocotyl instead of eight, and nine primary epicotyledonary bundles instead of six. The nine primary epicotyledonary bundles may not all divide, however, so that the number of bundles in the central region of the epicotyl is variable, ranging in general from fourteen to eighteen.

¹² While the material employed in this study traces its origin from individual plants, the possibility of hybridization in the field is not excluded. Thus any comparison which may be made in this place must be regarded as preliminary merely.

In both types of seedlings, but more frequently in the normal ones, additional or intercalary bundles appear in the hypocotyl, either *de novo* or as a result of division of the primary strands.

The following constants¹³ (table 19) for bundle number (at the different levels studied) epitomize the differences which characterize the two types of seedlings.

TABLE 19

	Trimerous Seedlings			Dimerous Seedlings		
	Mean	S. D.	C. V.	Mean	S. D.	C. V.
Root poles						
Minimum	5.02	.654	13.02	4.01	.081	2.03
Maximum	5.16	.739	14.47	4.13	.338	8.18
Mean	5.09	.707	13.87	4.05	.171	4.19
Primary double bundles						
Minimum	5.81	.288	4.86	4.02	.140	3.48
Maximum	5.98	.581	10.01	4.52	.666	14.74
Mean	5.91	.405	6.87	4.19	.411	9.66
Intercalary bundles						
Minimum09	.292	156.62	.07	.261	105.79
Maximum29	.686	381.67	.83	1.024	355.48
Mean19	.491	274.92	.49	.687	182.70
Mid-region of hypocotyl						
Minimum	11.99	.532	4.42	8.11	.409	5.04
Maximum	12.29	1.283	10.44	10.62	1.645	17.34
Mean	12.16	.883	7.24	9.23	1.193	12.67
Mid-region of epicotyl						
Minimum	14.89	1.152	7.74	12.11	.406	3.35
Maximum	16.10	1.750	10.87	12.36	.757	6.13
Mean	15.47	1.383	8.92	12.22	.586	4.79

The variability of root pole number is distinctly higher in trimerous than in dimerous seedlings, because of the fact that in *all* seedlings a four-poled condition is characteristic of the main root system and prevails even in the trimerous forms up to within a few millimeters of the base of the hypocotyl. Sections in the upper root region in such seedlings therefore show a considerable number of four- and five-bundled individuals.

The number of intercalary bundles is highly variable in both seedling types. The standard deviation is distinctly larger in the dimerous forms, but because of the generally lower average number of intercalary bundles in trimerous seedlings, the relative variabilities as measured by the coefficient of variation are higher in the trimerous type.

In the central region of the hypocotyl the variability of bundle number, both absolute and relative, is far higher in the dimerous seedlings, due in large part to the generally higher standard deviation of the number of intercalary bundles in the dimerous type.

In the central region of the epicotyl just the reverse is true, the variability of bundle number being higher in the trimerous than in the dimerous seedling. This is evidently due to the facts (*a*) that the intercalary bundles

¹³ Data for number of root poles are available for only three of the five lines.

of the hypocotyl are quite lost in the cotyledonary nodal vascular complex, and thus do not affect the variability of the dimerous plants; and (b) that the doubling of the primary epicotyledonary bundles which almost invariably occurs in the normal seedling may not always take place, at least not at as low a level as the central region of the epicotyl, in the abnormal type.

CONCLUSIONS

The results of the foregoing morphological and biometric analyses justify the emphasis at this point of certain general considerations.

1. External differentiation such as that which characterizes dimerous and trimerous seedlings of *Phaseolus vulgaris* is accompanied by profound differences in internal structure.

2. Anatomical characters are by no means constant. On the contrary, they are very variable even in series of individuals which are genetically highly homogeneous. Morphological investigations based on limited series of individuals may, therefore, result in inadequate conceptions.

3. Variation in anatomical structure is not constant for the plant as a whole, but may differ from region to region or from organ to organ. Thus in the regions of the seedling here under consideration, hypocotyl and epicotyl differ widely in the variability of bundle number. Furthermore, differences in variability from organ to organ or from region to region are not constant, but may be conditioned by other morphological features. To illustrate from the case in hand, the variability of bundle number of normal seedlings is higher in the hypocotyl than in the epicotyl. In seedlings with three cotyledons and three primordial leaves, just the reverse is true. These differences in biometric constants are readily understandable in the light of a knowledge of comparative morphology.

4. The results of this study emphasize the importance of the use of both biometric and comparative methods to supplement each other in any attack upon the problems of general morphology or of morphogenesis.